(12) INTERNATIONAL APPLICATION PUBLISHED UNDER THE PATENT COOPERATION TREATY (PCT)

(19) World Intellectual Property Organization
International Bureau

(43) International Publication Date
19 July 2001 (19.07.2001)

PCT

(10) International Publication Number
WO 01/50955 A1

(51) International Patent Classification[7]: A61B 5/05

(21) International Application Number: PCT/US01/01159

(22) International Filing Date: 12 January 2001 (12.01.2001)

(25) Filing Language: English

(26) Publication Language: English

(30) Priority Data:
09/483,163 14 January 2000 (14.01.2000) US

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(81) Designated States *(national)*: AL, AM, AT, AU, AZ, BA, BB, BG, BR, BY, CA, CH, CN, CU, CZ, DE, DK, EE, ES, FI, GB, GD, GE, GH, GM, HR, HU, ID, IL, IN, IS, JP, KE, KG, KP, KR, KZ, LC, LK, LR, LS, LT, LU, LV, MD, MG, MK, MN, MW, MX, NO, NZ, PL, PT, RO, RU, SD, SE, SG, SI, SK, SL, TJ, TM, TR, TT, UA, UG, US, UZ, VN, YU, ZA, ZW.

(84) Designated States *(regional)*: ARIPO patent (GH, GM, KE, LS, MW, MZ, SD, SL, SZ, TZ, UG, ZW), Eurasian patent (AM, AZ, BY, KG, KZ, MD, RU, TJ, TM), European patent (AT, BE, CH, CY, DE, DK, ES, FI, FR, GB, GR, IE, IT, LU, MC, NL, PT, SE, TR), OAPI patent (BF, BJ, CF, CG, CI, CM, GA, GN, GW, ML, MR, NE, SN, TD, TG).

Published:
— *with international search report*

For two-letter codes and other abbreviations, refer to the "Guidance Notes on Codes and Abbreviations" appearing at the beginning of each regular issue of the PCT Gazette.

WO 01/50955 A1

(54) Title: IMPROVED ENDOSCOPIC IMAGING AND TREATMENT OF ANATOMIC STRUCTURES

(57) Abstract: The present invention discloses methods of infrared endoscopic imaging technology and the uses thereof. Specifically, devices and methods that allow visualization of sensitive structures normally invisible under visible light illumination in real-time are presented. Examples of the imaging technology are confocal imaging, pulse oximetry, laser doppler and transillumination. Also provided are various configurations of the endoscopic devices.

IMPROVED ENDOSCOPIC IMAGING AND TREATMENT OF ANATOMIC STRUCTURES

BACKGROUND OF THE INVENTION

Field of the Invention

The present invention relates generally to the field of medicine. More specifically, the present invention relates to infrared endoscopic imaging technology and the uses thereof.

Description of the Related Art

Visualization of anatomical components that reside deep in body cavities, is a complex medical problem. Though endoscopy has revolutionized certain aspects of surgery on such components, certain risks are still apparent. Among these, the risk of accidentally cutting a vein or artery that lies near the treatment volume is very high, and places the patient's life in jeopardy. Such blood vessels may be invisible to the endoscopic

surgeon using regular (white light) illumination due to the relatively opaque nature of biological tissues.

Use of radiant energy to image anatomical structures that reside deep in tissues, or those that are obscured by tissues, has not been possible due to the excessive scatter and absorption of photons, and thus the relatively short mean-free-path between scattering and/or absorbing interactions. The consequence of this is that photons transmitted through, or reflected from, tissue and anatomic structures have suffered multiple scattering events and thus carry little or no image information at all and thus provide little useful information.

The brain depends on normal cerebrospinal fluid (CSF) flow for physiological and biomechanical homeostasis. Hydrocephalus results from an obstruction anywhere along the pathway of the flow. The mortality from this condition was extremely high until the 1950's when CSF diversion into the peritoneal cavity was introduced using rubber, plastic, and finally silastic shunts. However, the treatment of hydrocephalus is far from ideal, most procedures require craniotomy and procedures carry a 2% annual mortality and 10-30% morbidity with shunt obstruction, disconnection, overdrainage, and infection.

With the advent of narrow, high resolution endoscopes, third ventriculostomy for treatment of hydrocephalus can now be performed as a minimally invasive operation. Third ventriculostomy is an endoscopic procedure that involves producing a small perforation through the floor of the third ventricle whereupon circulation of the CSF can take

place. However, despite promising results from recent studies, the operation has failed to gain acceptance by the neurosurgical community in general because lying directly beneath the floor of the third ventricle is the terminal portion of the basilar artery with it's many important branches. Injudicious puncture of the floor can result in uncontrollable hemorrhage and death. A means with which to image sensitive structures through the floor of the third ventricle would lessen the risks and thereby increase the acceptance of this most valuable and proven operation.

While biological tissue is made up of an enormous number of different molecules, its absorption properties are dominated primarily by water, nucleic acids, and proteins (e.g hemoglobin and melanin). The optical properties of tissue are a function of wavelength, λ. Near-infrared (NIR; λ=700-1900 nm) radiant energy is the most penetrating in tissue. Furthermore, the scattering efficacy of tissue decreases with increasing wavelength, as predicted by Rayleigh and Mie scattering theory.

The ideal radiant energy for imaging the basilar artery during a third ventriculostomy would penetrate the unpigmented tissue that makes up the thin floor of the third ventricle, and also penetrate the CSF. On the other hand, the basilar artery, which contains blood, must be absorbing and/or scattering (with respect to the surrounding tissue and CSF) so it can be differentiated. Blood is contained within the artery, and the optical properties of blood are significantly different from those of soft tissue. This difference has been used in the development of transcutaneous blood-vessel viewers.

The ideal radiant energy for perforating the floor of the third ventricle would be highly absorbed in tissue and CSF (so as to limit damage to uninvolved structures), and yet would not produce problematic significant acoustic transients. Alternatively, formation of a laser-induced plasma at or near the floor of the third ventricle, using radiant energy of any wavelength, would also serve to fenestrate and yet minimize or eliminate propagation of radiant energy to nearby sensitive structures. The lasers should optimally be pulsed in order that a low average laser power could be used and that the produced heat would have little time to diffuse out to sensitive structures. On the other hand, the ideal radiant energy for coagulation of dissected tissues would be less strongly absorbed in tissue and would produce a coagulation effect without dissection.

The prior art is deficient in the lack of a device and methods that allow the neurosurgeon to visualize sensitive structures in real-time, thus providing important information and improved safety procedures. The present invention fulfills this long-standing need and desire in the art.

SUMMARY OF THE INVENTION

In one embodiment of the instant invention, a method of imaging deep anatomic structures normally invisible under white-light illumination is provided. This method comprises detecting infrared light from said region of interest

with an infrared sensitive image detector attached to an endoscope and a light source.

In another embodiment of the instant invention, the infrared sensitive image detector detects infrared light reflected or emitted from said region of interest. Examples include infrared sensitive video cameras and confocal imaging optical systems

Another embodiment of the instant invention is detection of infrared light absorbed by said region of interest to produce an image. Differences in the absorption spectra of myoglobin, HbO_2 and Hb proteins are especially useful for blood-tissue contrast, distinguishing arteries from veins, and detecting ischemia.

Another embodiment of the instant invention is the addition of one or more chromophores to the region of interest prior to imaging. These chromophores may enhance the image by adding contrast, fluorescing, and associating with specific tissues or organs. Examples of chromophore include calcium-linked dyes, iodine-linked agents, dye-tagged antibodies, and Indocyanine Green (ICG).

Yet another embodiment of the instant invention the region of interest is treated with δ-aminolevulinic acid to enhancing the production of porphyrin chromophores in malignant tissues. Another embodiment of the instant invention is transilluminating a region of interest by placing an infrared light source in contact with a tissue at or near said region of interest and detecting differences in infrared absorption or reflection properties to construct an image.

Another embodiment of the instant invention is illuminating a region of interest with different wavelength of light and detecting each wavelength independently for depth discrimination.

Other embodiments of instant invention entail methods of infrared image enhancement including Raman spectroscopy, multiphoton interaction, optical coherence tomography, time correlated single photon counting, optical rotatory dispersion, circular dichroism, polarization, chrono-coherent backscatter, simple inteferometry of backscattered light, and alignment of molecules in structures to be imaged by magnetic or electric fields.

Yet another embodiment of the instant invention is an endoscopic device capable of the real time imaging of subsurface structures normally invisible under white-light illumination. Such a device includes an endoscope, a light source, and an infrared sensitive image detector.

Other embodiment of the instant invention include infrared sensitive endoscopic devices with additional enhancements such as a laser doppler device to detect blood flow and quantify flow rates, a transilluminator and one or more infrared detectors, the ability to transmit light at different angles, and a means of cutting or puncturing tissue on the distal end of said endoscope.

Other and further aspects, features, and advantages of the present invention will be apparent from the following description of the presently preferred embodiments of the invention given for the purpose of disclosure.

BRIEF DESCRIPTION OF THE DRAWINGS

So that the matter in which the above-recited features, advantages and objects of the invention, as well as others which will become clear, are attained and can be understood in detail, more particular descriptions of the invention briefly summarized above may be had by reference to certain embodiments thereof which are illustrated in the appended drawings. These drawings form a part of the specification. It is to be noted, however, that the appended drawings illustrate preferred embodiments of the invention and therefore are not to be considered limiting in their scope.

Figure 1 depicts an endoscopic device showing imaging critical elements such as infrared (IR) filter and infrared sensitive CCD camera.

Figure 2A depicts a dual wavelength illumination device optionally employing an electrode to synchronize the video imaging with the pulse rate. **Figure 2B** depicts a method for imaging in multiple wavelengths whereupon optical filters are sequentially inserted into the image axis at a rate which is synchronized with the video capture rate.

Figure 3 depicts a confocal imaging device whereby white light illumination and optionally infrared illumination and/or white light illumination are used to image subsurface blood vessels.

Figure 4 depicts an imaging system employing Raman scattered photons and a scanner positioned in front of the spectrograph optical system or the infrared laser illumination.

Figure 5 depicts a blood vessel imaging device where motion detection is employed.

Figure 6 depicts a blood vessel imager where multiphoton effects are used for imaging.

Figure 7 depicts an optical coherence tomographic blood vessel imager.

Figures 8A-8F depict various ways in which polarized light and polarizing filters over the detector can be used to enhance blood vessel image contrast. **Figures 8C** through **8F** make use of an active device called a photoelastic modulator that can be used in the creation and analysis of polarized radiant energy.

Figure 9 is a diagram of an endoscopic device that is used to interrogate motion at the surface or beneath the tissue in a region of interest.

Figure 10 is a diagram of scattered, reflected photon paths in media with two different sets of optical interaction coefficients.

Figure 11 is a diagram of imaged photons that are collected with detectors of different degrees of collimation.

Figure 12 is a diagram of an initial radiant energy pulse saturating absorbers/scatterers, and the subsequent pulse that propagates further in the tissue than the initial pulse.

DETAILED DESCRIPTION OF THE INVENTION

Among the embodiments of the present invention, a method and device for visualizing such vessels, during surgery and in real time is provided. These embodiments can also be used to visualize other subsurface anatomic structures, such as muscle tissue. Though it is an object of these inventions to image tissues through an endoscope, these techniques may also be used externally to image structures, such as blood vessels, which lie beneath the skin.

Some preferred embodiments of the present invention overcome some of the limitations of using radiant energy for imaging tissues by altering the optical properties of the tissues themselves, thereby improving the ability to image by reducing scatter. Illumination methods, including transillumination, provide an optimal means for injecting photons into the region of interest. Additionally, some of the embodiments of the instant invention work by discriminating photons as a function of the average maximum depth to which they penetrated, or by virtue of their differing spectroscopic properties.

The instant invention presents a device and methods that allow the neurosurgeon to visualize these sensitive structures in real-time, thus providing important information that will improve the safety of the third ventriculostomy procedure. Also described is an optional design and method for delivering laser radiant energy to cut through the floor without damaging the structures underneath that could control

hemorrhage if it should occur and that would not affect the surrounding vital structures, such as the hypothalamus. This imaging/laser device could further be applied to other intracranial operations requiring accurate fenestration such as multi-loculated hydrocephalus, arachnoid cysts and even tumors. Finally, the technology will be applicable to other endoscopic surgical procedures in, for example, urology and gynaecology.

The most basic arrangement of the invention uses infrared endoscopic imaging technology for the purpose of visualizing subsurface structures that are normally invisible under white-light illumination. In one embodiment, a standard endoscope and coupled white-light source, already commercially available is used (Figure 1). The endoscopes, when used for neurosurgery, for example, typically employ a charge-coupled device (CCD) color video camera in the proximal end, the output signal of which is captured by a video display monitor. The surgeon then manipulates the endoscope by looking at the monitor. In the present invention, the color video camera is replaced with either an infrared (IR) sensitive monochrome video camera, a color video camera, or an infrared (IR) sensitive monochrome video camera which images sequentially through red, green, blue and infrared filters so an IR and color image can be simultaneously formed, or a color camera that is also sensitive to IR radiation and which the user selects to image either through an open aperture to give an unaltered color image, or through an IR filter. It may be necessary to transmit the image of the region-of-interest (ROI) through a coherent

optical fiber bundle, or other optical image guide, to the imaging equipment which, due to it's bulky nature, is positioned on a nearby stand and not on the proximal end of the endoscope.

It is optional whether the surgeon would want the IR and color image combined and displayed on the same monitor; i.e., the IR information could undergo thresholding such that pixel intensity values over or under a particular value are retained and all other pixel values are rejected thus allowing one to select, for example, a strongly IR absorbing blood vessel alone out of the entire IR image, or whether to have the IR image displayed on a dedicated monochrome video monitor adjacent to the color video monitor which displays the typical white-light color image.

All of the refinements described herein are intended to be used in conjunction with the invention described above; i.e., to image a ROI wholly or in part with an infrared sensitive image detector coupled to an endoscope.

The current invention is directed to a method of imaging deep anatomic structures normally invisible under white-light illumination. This method comprising inserting an endoscope into an anatomical region of interest while illuminating said region of interest with a light source and detecting infrared light from said region of interest with an infrared sensitive image detector. The resulting images may be displayed on a video monitor.

The current invention is directed to methods where in the infrared sensitive image detector detects infrared light

reflected or emitted from said region of interest. Examples of methods of detecting reflected or emitted infrared light include an infrared sensitive monochrome video camera, a color video camera which images sequentially through red, green, blue and infrared filters, an infrared sensitive monochrome video camera which images sequentially through red, green, blue and infrared filters, and a color camera which is also sensitive to infrared radiation which can alternatively image through an open aperture or an infrared filter.

The current invention is also directed to methods in which the a confocal imaging optical system is used to detect infrared reflections and emissions.

The instant invention is also directed to a method wherein the light source is a laser which emits near infrared light of wavelengths similar to the dimensions or textures of objects being imaged. As a result, reflection or said light results in detection of a speckle pattern. Movement or the speckle pattern can be used to detect red blood cell movement and blood vessels.

The instant invention is also directed to methods by which infrared light absorbed by said region of interest is imaged. Differences in the absorption spectra of myoglobin, HbO_2 and Hb proteins can be used localize each protein in an image. Such imaging proves useful in differentiating the location of Hb and HbO2 contained within vessels from Mb to produce good blood-tissue contrast, differentiating the location of Mb and Hb/HbO2 to distinguish arteries from veins,

measuring the relative amounts of Hb, HbO2 and Mb to provide to detect ischemia.

The current invention is also directed to a method by which red and near infrared light are emitted by the light source and the infrared sensitive image detector measures the absorbance of the red and infrared light.

The instant invention is also directed to a method wherein one or more chromophores are added to said region of interest. These chromophores may enhance the image by adding contrast, fluorescing, and associating with specific tissues or organs. Examples of chromophore include calcium-linked dyes, iodine-linked agents, dye-tagged antibodies, and Indocyanine Green (ICG).

The instant invention is also directed to a method by which a region of interest is treated with δ-aminolevulinic acid to enhancing the production of porphyrins in any malignant tissues. These porphyrins then act as chromophores during imaging.

The current invention is also directed to a method of transilluminating a region of interest by placing an infrared light source in contact with a tissue at or near said region of interest. The light is scattered through the region of interest, and infrared detectors are used to detect light reflected or absorbed during transillumination. Differences infrared absorption or reflection properties are then used to construct an image.

The instant invention is also directed to a method of obtaining depth discriminate information in an image by

illuminating a region of interest with different wavelength of light and detecting each wavelength independently.

The instant invention also includes a method by which the average range of tissue penetration of detected photons can be selected by changes in the degree of detector collimation. Detector collimation can be selected by a physical collimator of a specific dimension and shape, a narrow-bandpass interferometric filter, light-absorbing baffles between objective element and detector, and confocal techniques.

The instant invention is also directed to a method of enhancing an image by saturating absorbing sites in aregion of interest with a strong pulse of radiant energy and applying additional pulses of radiant energy before the effects of prior pulses have diminished.

The instant invention is also directed to other method of infra red image enhancement including Raman spectroscopy, multiphoton interaction, optical coherence tomography, time correlated single photon counting, optical rotatory dispersion, circular dichroism, polarization, chrono-coherent backscatter, simple inteferometry of backscattered light, and alignment of molecules in structures to be imaged by magnetic or electric fields.

The instant invention is also directed to an endoscopic device capable of the real time imaging subsurface structures normally invisible under white-light illumination. Such a device includes an endoscope, a light source, and an infrared sensitive image detector. Examples of infrared sensitive image include an infrared (IR) sensitive monochrome video

camera, a color video camera which images sequentially through red, green, blue and infrared filters, an infrared (IR) sensitive monochrome video camera which images sequentially through red, green, blue and infrared filters, and a color camera which is also sensitive to IR radiation which can alternatively image through an open aperture or an IR filter. The infrared sensitive image detector is placed on the proximal end of said endoscope or the image may be transmitted to an infrared sensitive image detector through an image guide such as a a coherent optical fiber bundle .

The instant invention is also directed to infra red sensitive endoscopic devices with additional enhancements such as a laser doppler device to detect blood flow and quantify flow rates, a transilluminator and one or more infrared detectors, the ability to transmit light at different angles, and a means of cutting or puncturing tissue on the distal end of said endoscope.

The following examples are given for the purpose of illustrating various embodiments of the invention and are not meant to limit the present invention in any fashion.

EXAMPLE 1

Imaging techniques

Pulsatile enhanced imaging

Pulse oximeters are devices that measure the percent oxygen saturation of blood by non-invasive optical means. This

is accomplished by passing red ($\lambda_1 \approx 660$ nm) and near infrared ($\lambda_2 \approx 940$ nm) radiant energy through a layer of tissue, typically a fingertip. Based on the relative signals transmitted through the fingertip at each wavelength, and knowing the absorption characteristics of the pertinent absorbing chromophores (tissue, oxygenated hemglobin or HbO_2, and deoxygenated hemoglobin or Hb), and after extensive calibration of the device with a direct measurement of blood oxygenation, it is possible to obtain the percent oxygen saturation of the blood. One important operating characteristic that makes the pulse-oximeters work is that they can discern between arterial and non-arterial absorption by disciminating between time-varying signals due to pulsatile flow because of the pumping heart; an effect more evident in arterial blood and steady-state signals from venous blood and tissue. This methodology of differentiating blood and/or arterial blood from tissue has not been used for imaging.

Figure 2(a) shows a device; e.g, video camera, imaging the surface of an anatomic structure. The illuminating light consists of alternating pulses of radiant energy produced by light-emitting diodes (LEDs) filtered with a bandpass filter or diode lasers, to impinge on the tissue with a wavelength of, for example, λ_1=660 and λ_1=940 nm. The detector; e.g., charge-coupled device, or CCD, captures an image of the tissue every $1/30^{th}$ of a second, and so the illuminating light is alternately pulsed once every $1/30^{th}$ second. The images captured can be digitized and analyzed mathematically in a way similar to standard transillumination pulse-oximeters, except in this case reflectance, R, is determined, and not transmittance, T. The

mathematical analysis is none-the-less similar considering that optical density, OD, is related to R by OD=-Log(R) and T=1-R. In a fashion similar to pulse oximetry, the images can be decomposed into an time-varying signal, a.c., and steady state,d.c., signal, whereby the former is due to absorption within arteries and to a much lesser degree, veins, while the latter is primarily due to non-arterial absorption. Alternatively, an electrocardiogram (ECG) electrode could be used to monitor the heartbeat in order to match the phase of the signal with the heartbeat, thus achieving the same aforementioned result. This methodology would allow one to optically differentiate arterial blood absorption from absorption due to other biomolecular species, such as deoxygenated blood. This can therefore be used to enhance the contrast between blood vessels and the surrounding tissue. Furthermore, it optionally measures blood oxygen saturation, which when combined with the imaging information, provides useful diagnostic information regarding the spatial distribution of oxyhemoglobin, or ischemia, a lack of sufficient oxygenation in tissue.

Alternatively, a rotating filter wheel in front of the camera (Figure 2b) and a single illumination source that is filtered to produce broadband radiant energy that includes the necessary wavelengths, λ_1 and λ_2 is used. The rotation rate and phase, with respect to the detector, of the optical filter wheel is adjusted to maximize the a.c. signal and thus is in phase with the pulsatile arterial flow, or is controlled by a pulse rate signal from an optional ECG electrode.

The use of multiple wavelengths of radiant energy to achieve imaging contrast are similarly used to provide contrast between veins and arteries and/or hemoglobin within blood vessels versus extravascular globins. Myoglobin (Mb) is a protein similar to Hb that transports oxygen in muscles and which provides much of the visual appearance of color in muscles. The absorption spectrum of myoglobin is different from HbO_2 and Hb. This difference is used to differentiate Hb and HbO_2 contained within vessels from Mb, thus producing good blood-tissue contrast. Similarly, the difference in the absorption spectrum of Mb and Hb/HbO2 is used to differentiate arteries from veins insofar as arteries have a layer of smooth muscle cells containing Mb while veins do not. Using the relative measurements of Hb, HbO_2 and Mb also provides a sensitive means with which to measure ischemia.

A measurement of Mb in bulk tissue is useful in itself in that it gives an indication of tissue ischemia. For example, in laser myocardial reperfusion, repair to ischemic heart tissue is made by producing channels within the muscle wall of the heart through to the inner chamber so that oxygenated blood can flow into the oxygen starved muscle. Visualizing the magnitude of Mb radiant energy absorption allows the cardiac surgeon to determine the physical extent, and the magnitude of the tissue ischemia. This is beneficial in that the surgeon is able to localize the laser produced channels so that the tissue in need of oxygenation is treated, while healthy tissue remains undamaged by laser irradiation. It is a further object of the present invention to provide a means to visualize myoglobin and

hemoglobin rich tissues, and those tissues rich in oxygenated derivatives, in order to perform such reperfusion procedures.

Confocal enhanced imaging

The concept of rejecting light, scattered from locations other than the point being imaged, namely, with coordinates x,y,z, by using apertures in the imaging system, is referred to as confocal imaging. This concept has been used for the last decade or so in providing a means with which to "optically section" microscopic specimens being viewed with a microscope. Confocal microscopy normally uses white light illumination, or ultraviolet (UV) or Argon-laser or Argon-krypton illumination to induce fluorescence in the sample. The former illumination results in significant chromatic aberrations in the final image, while the latter provides only a fluorescent image.

It is beneficial, in certain cases, to use red and/or near infrared radiant energy in a confocal imaging optical system. Narrow band illumination through the use of diode lasers or bandpass filtered broadband light sources is best utilized (Figure 3). By alternately using, for example, 660 nm and 940 nm radiant energy in a confocal imaging system, it is possible to collect information on HbO_2 and Hb as a function of depth in the sample, thereby allowing for optimal visualization of blood vessels such as those below other intervening structures.

Raman enhanced imaging

Raman scattering is an inelastic interaction between photons and molecules. The photons scattered from an interaction between a photon and molecule do, to a small degree, have slightly less or slightly more energy per photon than the incident photon. These scattered photons have been utilized to obtain infrared spectral information of matter. These scattered photons can be used for imaging information.

Raman spectroscopy is a light scattering technique that typically uses laser radiation to excite the sample whereby the scattered radiation emitted by the sample is analyzed. Emission data has two main characteristics: (1) the frequencies at which the sample emits the radiation (a small number of the incident photons, perhaps only 1 in 10^6, is emitted at frequencies different from that of the incident light), and (2) the intensities of the emissions. Determining the frequencies allows identification of the sample's molecular makeup, since chemical functional groups are known to emit specific frequencies and emission intensity is related to the amount of the analyte present. With highly sensitive charge-coupled-device (CCD) and efficient optics such as transmission holographic gratings and notch filters, Raman spectroscopy has enormous potential in the medical field.

It is useful to illuminate the anatomic structure of interest with the radiant energy produced by a 850 nm diode laser, and a Raman spectrograph for imaging (Figure 4). The Raman scattered photons specific to blood then are used to detect and discriminate blood from other biomolecules. The

same idea is extended to other biomolecular species of interest such as circulating pharmaceuticals, Mb and glucose. To build up an image using Raman scattered photons, it may be necessary to either scan the illumination system in a raster-scan, for example, and capture information point-by-point before a 2-dimensional image can be made up. Alternatively, the detector can be scanned. Furthermore, a 2-dimensional imaging detector, such as a CCD, can be used. The strong molecular specificity of Raman scattering allows for good rejection of signals not pertaining to the molecule of interest.

Laser speckle enhanced imaging

If the movement and/or texture of objects reflecting coherent laser radiant energy have dimensions on the order of the wavelength of the radiant energy, then constructive and destructive interference can take place. To the eye, this often gives the appearance of a "speckle" pattern superimposed on the illuminated object. If the object reflecting the laser light moves, then the speckle pattern moves. Such movement can be used to detect motion. This concept is used to detect blood flow. For example, Figure 5 shows an imaging system being used to detect blood within a vessel. The imaged vessel and surrounding structures are illuminated with radiant energy produced by a laser, preferably in the near infrared region of the spectrum, so relatively deep penetration of the radiant energy occurs, and yet is reflected from red blood cells (RBCs). Any changing speckle pattern is a consequence of RBC movement. The reflected radiant energy is captured by a

detector, which is positioned behind an aperture. Thus, any change in the speckle pattern results in a change in the detector's signal output. If the infrared laser is scanned, then an image of the speckle pattern builds up and RBC movement is detected, thus providing a means with which to detect blood vessels.

Multiphoton interaction enhanced imaging

Under certain irradiation conditions, it is possible to have photons, with a wavelength λ, interact with an atom or molecule which normally would not absorb at λ, but which do absorb at $1/2\lambda$. For example, fluorescein with an absorption peak around 500 nm is induced to fluoresce when irradiated with two photons with a wavelength of 1000 nm. Three-photon excitation can also be used in certain circumstances. This multi-photon effect requires lasers which produce high peak powers; e.g. >2kW in a pulse length, τ_p<1 ps, and yet have a low average power so that there are no undesirable laser-tissue interactions such as photothermal coagulation. This multi-photon effect is used for fluorescent imaging as the requisite high photon density is made to occur only at the focus of the laser beam, which then is scanned in three dimensions. The benefits of multi-photon excitation is that the incident radiant energy is not attenuated by absorption of the fluorochrome above the plane of focus and the longer excitation wavelength used are less Raleigh scattered.

It is generally believed that multi-photon techniques are useful only for fluorescence imaging. However, it is known

that in a classical physics description, the scattering and absorption effects of tissue are mathematically related to each other. Thus, in parts of the electromagnetic spectrum where tissue is highly absorbing, it is also highly scattering. Thus, a multiphoton effect can be used to gather imaging information. For example, consider that blood absorbs strongly at about 400-425 nm, however radiant energy with this wavelength, which appears blue, is strongly absorbed in tissue and so only penetrates superficially. However, it is possible with two-photon scattering and absorption to obtain information about blood using radiant energy at 800-850 nm. Such radiant energy is quite penetrating in tissue, and yet will interact with blood if the photon density is large enough. It is therefore possible to obtain imaging information (Figure 6) using a pulsed laser producing 850 nm radiant energy. For practical reasons, such an imaging scheme may benefit from using a Q-switched Nd:YAG laser (1064nm), as these lasers are relatively inexpensive and fortuitously blood absorbs strongly at 532 nm. The 532 nm scattered information is collected in synchrony with the pulsed Nd:YAG laser. On alternate scans, white light or infrared images is captured. Comparison of the two could be used to determine the location of the blood or other light absorbing/scattering chromophore in the field of view.

Optical coherence tomography enhanced imaging

Optical coherence tomography (OCT) is based on low-coherence interferometry; e.g. it can employ a white-light Michelson interferometer. High resolution depth-dependent

imaging is obtained by focusing light from an optical low coherence source on the biological tissue and interfering the backscattered intensities with the incident light. Two dimensional images are obtained by performing the interferometric measurement as a function of axial and transverse position on the tissue. Depth information in the tissue is collected by varying the reference arm pathlength in the interferometer. Useful interferometric information occurs only when the optical pathlengths of the light traversing the reference path and tissue path are identical to within the coherence length of the source. OCT is beneficial in that it does not require an illumination source with a long coherence length; e.g., a laser and can be done with the use of optical fibers.

It is benefical to use OCT with infrared radiant energy to obtain good penetration of the light (Figure 7). By using several wavelengths of light sequentially, as is done in pulse oximetry, specific images of arteries and veins could be obtained, as could a non-invasive measurement of blood oxygen saturation in a particular imaged vessel.

Time correlated single photon counting enhanced imaging

Time-correlated single-photon counting (TCSPC) is a statistical technique which may be used to measure the time profile of the emission of a sample following excitation by a short light pulse. The time delay between a trigger ("start") pulse, which is fixed in time with respect to the excitation pulse, and the moment of arrival of a photon emitted by the sample and then detected by a photomultiplier ("stop" pulse) is

recorded. By accumulating many such intervals in a histogram, the probability that a photon is emitted by the sample at a certain moment is measured, i.e., the time profile of the emission is measured.

TCSPC is a commonly used technique in fluorescence spectroscopy due to its wide dynamic range, high time resolution and high sensitivity. Time resolutions of the order of 50 ps (FWHM) are easily achievable with commercial ultrafast laser and detector systems. Typically, a mode-locked argon-ion laser is used to synchronously pump a cavity-dumped dye laser. The resultant train of picosecond pulses is coupled into an optical fiber in contact with the sample under investigation. The pulse shape changes as it propagates through the medium. This is due to the fact that photon pathlengths are altered through interaction, such as scattering and absorbing, with the medium. In fact, the optical properties of the medium are inferred from the shape of the emitted pulses. Typically, the emitted pulses are collected with an optical fiber positioned on the sample at a known distance from the input fiber. The distal end of the collection fiber is in contact with the detector shich, for ultrafast applications, is a microchannel plate–photomulitplier tube (MCP-PMT).

The optical properties, such as scattering and absorption, of the material are obtained through non-linear least squares fitting of the data (amplitude vs. time spectra) to a model function. A diffusion model is commonly used to fit the data. Analytic solutions of the diffusion equation in homogeneous media exist for various simple geometries. This

data, the absorption and scattering coefficients, can be collected during scanning in the x-y plane in order to build up an image.

Optical rotatory dispersion, circular dichroism and polarization enhanced imaging

The optical behavior of some biomolecules depends on the state of polarization of the incident photons (D. Freifelder, *Physical Biochemistry-Applications to Biochemistry and Molecular Biology*, W.H. Freeman and Co., NY, 1982). Such a molecule is called optically active in that it exhibits different indices of refraction and different molar absorption coefficients for left and right-hand circularly polarized light. DNA, for example, is optically active. Optical activity can be detected using the methods of optical rotatory dispersion (ORD) spectroscopy or by circular dichroism (CD) spectroscopy. The strength of the ORD or CD spectra, which depend on wavelength, but are typically done in the UV region of the spectrum between about 190 and 300 nm, are dependent on the particular molecular specie.

Circular dichroism (CD) and optical rotatory dispersion (ORD) are difference measurements in that CD measures a difference between the absorption in matter of radiant energy that is left and right circularly polarized light, while ORD measures the difference in the refractive indices for left and right circularly polarized light. If a biomolecule is optically active that is dissymmetric and non-superimposable mirror images of the molecule occur, then circularly polarized light interacts with the molecule depending on the handedness

of the light. Molecular shape and orientation also determine the degree to which linearly polaried light interacts with a molecule. For example, polarizing sunglasses have absorbing chromophores that are mostly oriented horizontally; thus when light specularly reflected from a roadway, which is, to a degree, linearly polarized with the axis of polarization oriented horizontally, then it is absorbed by the chromophores. Unpolarized light retroreflected from a tissue interface will have a degree of linear polarization; incident linearly polarized light retroreflected (Figure 8a) from deeper within the tissue will loose the initial polarization, depending on how many scattering events occur within the tissue.

These ideas can be used to improve the ability to image subsurface anatomic structures. For example, specular reflectance from tissue interfaces, which tend to obscure the image of a subsurface structure, can be reduced or eliminated by linearly polarizing the incident light, and incorporating a linear analyzer whose transmission axis is oriented orthogonal to the transmission axis of the incident light polarizer. Similarly, by passing unpolarized light through a linear polarizer and 1/4 wave retarder, circularly polarized light is produced which, when retroreflected, will be absorbed upon passing through the same 1/4 wave retarder and linear polarizer (Figure 8b). Note that these polarization techniques which serve to reduce unwanted specular reflection also are used to increase the ratio of photons scattered a few times (superficially penetrating photons) to photons scattered many times (deeper penetrating photons). This provides a way to

either reject highly scattered photons that degrade image contrast or to acquire depth discriminate information.

These same methods are used for molecular discrimination. Devices that measure the reflectance and absorbance from tissue of polarized light and measure the state of polarization in the reflected light can make use of a photoelastic modulator (PEM). This device, depending on the waveform and phase driving it, is used to analyze polarized light when used in conjunction with polarizers and waveplates. The frequency at which the PEM is driven and the frequency of the signal detected also play a role in determining what exactly is being measured (Hinds Instruments, Inc., Oregon).

For example, optically active molecules such as glucose can be detected by transmission measurements using CD or ORD. PM-IRRAS strands for Polarization Modulation Infrared Reflection-Absorption Spectroscopy. It is the differential IR absorption between the s- and p- linearly polarized light for the molecules in a tissue (Figure 8C). In ellipsometry, the polarization change of a light beam is measured when it is reflected by the sample. This change in polarization is then related to the sample's properties (Figure 8D). Vibrational circular dichroism (VCD) is the differential absorption between left and right circularly polarized light. It is a measurement of the optical activity for chiral molecules (Figure 8E). Linear dichroism (LD) is the differential absorption between two orthoganal, linearly polarized states. LD is a measurement of the sample's bulk property that is a result of the regular orientation of the molecules in the sample (Figure 8F with the

PEM set at 0 egress and phase-sensitive detector at the 2nd harmonic).

Laser Doppler enhanced imaging

Laser doppler devices are well known in the medical device arena. They are useful for detecting blood flow and for quantifying flow rates. Laser Doppler measurements can be done through thin optical fibers, which can be inserted down working channels in an endoscope. For the purpose of sensing subsurface vasculature, a laser Doppler device is incorporated into the system whereupon the surgeon gains information on the presence of a nearby vessel by virture of the increased laser-Doppler signal.

EXAMPLE 2

Methods to enhance imaging techniques

The following processes are optionally used to enhance the imaging techniques.

Image processing

Image processing devices, which manipulate digital image data quickly, enhance any of the imaging techniques discussed. It is an object of the present invention to process the image such that the illumination, as projected or detected, is equally distributed across the ROI. This illumination compensation improves the quality of the image obtained by reducing uneven lighting of the ROI. Image processing devices

that perform such enhancements are commercially available. For example, Dage-MTI (Michigan City, IN) markets a device that can perform real-time edge enhancement, uneven illumination compensation and frame averaging.

Vascular contrast enhancement

In order to improve the imaging contrast between blood vessels, or other sub-surface structures, and the surrounding tissue, exogenous chromophores are used. For example, Indocyanine Green (ICG) is a well know agent used for many years in quantifying cardiac output and imaging retinal vasculature. When in plasma, it has an absorption maxima at about 800 nm. By employing multiple-wavelength illumination and/or detection whereby one of the images captured is illuminated or detected around 800 nm, and the other at a wavelength where ICG is only weakly absorbing at, for example, 660 nm, then it is possible to differentially image vasculature, as is already known. However, when combined with the aforementioned pulsatile imaging methodology, further contrast enhancement is achieved. This same idea of using exogenous chromophores can be extended with a range of other molecules, some of which have a beneficial tendency of concentrating in diseased tissue. For example, it is know that δ-aminolevulinic acid (δ-ALA) collects in malignant tissue where it greatly enhances the production of porphyrins. These porphyrins are strongly absorbing and fluorescent thereby providing a technique to identify and treat cancer.

In the case of subsurface blood vessels, some form of motion detection combined with the imaging technology is beneficial. For example, by splitting the signal reflected from the region-of-interest, and directing one of the split signals into a CCD detector, which monitors movement by monitoring the change in signal at each pixel as a function of time (Figure 9), it is possible to superimpose on the regular infrared image information on movement. This allows a surgeon to detect a subsurface blood vessel by virtue of the movement induced by the vessel in the surface of the imaged tissue.

Photon injection geometry also plays an important role in contrast enhancement. An improved image is obtained when incident light penetrates the tissue and interrogates the object from an angle other than perpendicularly relative to the object.

Transillumination

When a light emitting device is placed in contact with a tissue, the photons injected into the tissue are scattered. During this process of transillumination, many of these photons will encounter objects below the tissues and be absorbed and/or reflected by the objects. Because the light encountering the objects has been scattered, it arrives at the object from below or from the side of the object, as well as from above. Light which escapes the tissues following such encounters contains information that can be used for imaging. Use of infrared sensitive detectors with infrared containing light sources as transillumination devices results in the ability to detect subsurface structures that are differentiated on the basis of their

infrared absorption or reflection properties. For example, in the instant invention, an infrared endoscopic illuminator placed in contact with a tissue will inject photons into the tissue that illuminate objects in proximity to the site of injection. In this case, blood vessels are detected by an infrared sensitive detector that collects photons emerging from the tissues following scattering events in the tissues.

Alternating color and infrared imaging

Most surgeons are most familiar with endoscopy using color sensitive cameras. Color information is used by the surgeon as diagnostic information. On the other hand, infrared sensitive cameras only produce a black-and-white image. It is beneficial to devise an imaging system whereby the surgeon can either select to quickly change from color to B&W imaging, or to collect the most relevant infrared image information; e.g., the location of a subsurface blood vessel, and to superimpose this information on the color image. Another way to achieve the same result is to use an infrared sensitive CCD detector in front of which is positioned a rotating filter wheel with red, green, blue and infrared bandpass filters. The filters are positioned in front of the CCD at a known rate and in synchrony with the capture of information from the CCD. The information is processed into an IR image and into a color image by combining the red, green and blue information.

Combining with therapeutic laser energy delivery devices

Optical fibers are used to guide the radiant energy of a laser down an endoscope, whereupon it is used to cut, coagulate, or induce fluorescence in tissue. It is beneficial in certain surgeries, such as third ventriculostomy, to be able to use infrared imaging to identify sensitive subsurface structures such as blood vessels, and then to use the radiant energy produced by a laser, such as the 2.94 micron wavelength radiation produced by an Er:YAG laser, to produce a fenestration in the membrane floor of the third ventricle.

Coherent imaging fiber bundle

Some preferred embodiments of the present invention are too bulky to mount on the proximal end of an endoscope. It might therefore, in certain cases, be beneficial to optically guide the image from the endoscope over to the image collection and processing device, which is positioned on a table nearby. Coherent imaging fiber bundles are suitable for this purpose. The input end of the bundle is positioned in the image plane of the endoscope, and the output end of the bundle is positioned in the object plane of the image collection system. This arrangement is light and flexible, and so gives the surgeon the necessary freedom-of-movement.

EXAMPLE 3

Depth Discriminationa; Depth Imaging Schema

Depth discriminate information is obtained using injected photons of differing wavelengths (Figure 10). It is well known, for example, that radiant energy with a wavelength λ_1 of 1064 nm, for example, has a much larger mean-free-path $\bar{z}_1$ in tissue than radiant energy with a wavelength λ_2 of 488 nm (mean-free-path $\bar{z}_2$), for example. Thus, by injecting beams of different wavelengths, and by detecting each separately by, for example, using the previously said techniques, or by using alternating band-pass filters with a center passband wavelength of λ_2 and λ_1 in front of the detector, depth discriminate information can be obtained. This concept is extended to photons of differing states of polarization since the optical properties of tissue depend, to a degree, on the state of polarization of the incident photons.

The degree of detector collimation can affect how photons with a particular average depth of penetration, are collected. For example (Figure 11), a strongly collimated detector at distance h from the surface of the tissue can be configured to collect photons which have propagated, on the average, through a particular region-of-interest (ROI). A less strongly collimated detector would collect photons that are less likely to have propagated only through the ROI. Capturing the data and comparing the two sets of data allows one to focus on the ROI and reject photons that carry information about the rest of the tissue. By increasing h, more specific photon discrimination

results due to a small solid angle being subtended by the detector, albeit at the expense of capturing less total number of photons.

Collimation is provided in a number of ways. Collimation by a physical collimator of a specific dimension and shape (e.g. a long, narrow tube) is one way, while a narrow-bandpass interferometric filter is another. In the latter case, any photons at the wavelength of the center of the passband, but that impinge on the filter element at non-normal incidence, will not be transmitted. Optionally, optical elements that image a particular small area of the skin onto the detector, and which have light-absorbing baffles between the objective element and detector for absorbing off-axis light are used. Confocal techniques, which strongly reject photons that arise from tissue that is out of the volume of interest, are used for photon discrimination. They have been used in microscopy, but not in photon discrimination of macroscopic objects such as blood vessels.

Raman scattering

The Raman spectra of hemoglobin and human breast tissue are different (H. Mantsch and D. Chapman, *Infrared Spectroscopy of Biomolecules*, Wiley-Liss:NY, 1996). Hemoglobin has absorption peaks at approximately 1082, 1204, 1236 and 1337 cm^{-1} which do not appear, or appear with reduced magnitude, in the spectra of breast tissue. This spectral information would be nearly impossible to detect *in vivo* using standard non-Raman infrared spectroscopy as the radiant energy source; it is so strongly absorbed in tissue that

reflectance or transmission spectra of deep structures would be impossible to obtain under normal conditions. Raman spectrometers can use incident radiant energy which is not strongly absorbed in tissue; e.g., 1064 nm radiant energy produced by an Nd:YAG laser, thus spectra are obtained to a clinically useful depth in tissue. By taking spectra at various sites in the x-y plane of tissue, and by taking the difference between the spectra at the relevent wavelengths, hemoglobin, for example, can be preferentially detected. This provides a way to image subcutaneous structures, such as blood vessels, or structures which are rich in blood. The schema outlined in Figures 1-6 demonstrate how to retrieve depth information. The use of incident beams with two different wavelengths is especially useful as the Raman spectra does not depend on the wavelength of the incident radiant energy, but the depth to which the incident photons penetrate in tissue most certainly is a function of wavelength. Thus, the only difference between two spectra obtained at the same x-y coordinates, but using two different wavelengths of incident radiant energy; e.g., 1064 nm and 633 nm, arise from the different average depths to which the photons propagated before they were Raman scattered.

Optical activity

While ORD and CD techniques have been used to obtain spectral information, *in vitro*, of biomoleuclar species, they have not been used *in vivo* for imaging. By irradiating tissue with radiant energy of alternating circular handedness;

e.g., photoacoustic modulators and polarizers, or alternating planes of linear polarization, and by detecting the polarization state of the reflected radiant energy, it is possible to discriminate: (1) photons which have traveled to particular average depths in tissue and (2) photons which have interacted with a particular optically active molecular species. ORD is advantageous over CD in this situation since the former extends far from absorption bands into spectral regions where the molecule is transparent, like infrared, and where the mean-free path in tissue is relatively large.

Interference Techniques

By combining incident and reflected photons in an interferometer, it is possible to discriminate between photons which have propagated different distances and, therefore, different average depths, in tissue. Techniques, such as chrono-coherent backscatter or simple inteferometry of backscattered light using interferometers such as can be constructed from optical fibers or mirrors and lenses, provide a way to depth discriminate. These methods are based on the fact that photons that have propagated a relative long distance in tissue do not tend to constructively interfere with the incident photons since their physical properites; e.g., phase, bear little similarity due to multiple scattering. By using interferometric techniques at a matrix of points in the x-y tissue plane, and by varying the reference arm in the interferometer, it is possible to obtain structural information as a function of depth. Optical coherence

tomography is such an interferometric technique suitable for imaging subsurface structures.

Doppler Techniques

Laser doppler flowmeters are sensitive to blood flow by virtue of photons being scattered off red blood cells. By raster scanning the interrogating laser beam and imaging optics over the surface of the skin, a two-dimensional image of blood flow is built up. This image is useful for determining the location of a blood vessel below the skin surface.

EXAMPLE 4

Depth Discrimination: Methods of altering tissue optical properties

Various methods of altering tissue optical properties are optionally useful in providing radiant energy for therapeutic uses to tissues. For example, to increase the depth of activating light penetration in photodynamic therapy, a pulse radiation protocol as described increases the depth of treatment. Similarly, in photothermal therapy pulsed irradiation is useful in increasing the volume of treated tissue and the depth to which treatment occurs. In addition, photons from continuous wave irradiation would also behave in an analogous manner.

Saturation

Given the normally fixed mean-free path in tissue of photons of a particular wavelength, altering the optical properties provides a means with which to obtain depth

discriminate information. Figure 12 shows an invention wherein absorbing sites in tissue are saturated with a strong pulse of radiant energy which is absorbed. While the lifetimes of excited molecular states in dense media such as tissue can be quite short (less than nanoseconds), if a second pulse of radiant energy is injected into the tissue before most of the sites have had time to relax to a lower energy state, then the tissue exhibits an increased mean-free-path to the subsequent pulse. Consequently, by sequentially capturing reflected photons from each alternating pulse and comparing the signals, the only difference in the signals is due to the fact that the photons in the second pulse propagated further in the tissue than the photons in the initial pulse. Thus, depth discrimination is possible.

Dyes

Image enhancing agents may also be added to the system. These are small molecular weight dyes that are infused into the system thereby providing contrast or by fluorescing, thereby providing image information about the structure in which they are located. In one embodiment, a contrast agent is added to the system that has optical properties very different from the tissue in which it sequesters, thereby increasing the amount of scatter or absorption within the system and thereby altering the probability of photons escaping the system and being detected. Alternatively, dyes alone or linked to certain substances may preferentially be retained by organs or tissues. Calcium-linked dyes may sequester in the bone, iodine-linked

agents often wind up in the thyroid, indocyanine green is retained in intact blood vessels, and dye-tagged antibodies will collect in tissues carrying specified antigens such as malignant or infected tissue. Thus, imaging of the ROI in tissue can be obtained using the aforementioned schema and contrast improving dyes or molecules.

Magneto- and Electro-Optical Effects

Use of magnetic or electric fields to align molecules in structures to be imaged provides a means with which to obtain contrast in a way not unlike how magnetic resonance imaging uses magnetic fields to induce rotations of nuclear magnetic moments in protons. Strong electromagnetic fields align molecules in such a way that the optical properties of the tissue are altered, especially the polarization-dependent optical properties. Alignment of molecular structures are x, y, z-coordinate dependent as determined by the non-uniformity of the electromagnetic field. Photons encountering aligned molecules in a medium or tissue will tend to interact or not interact, depending on orientation of the molecules with respect to the polarization state of the photons.

The Faraday effect involves rotating the plane of linear polarization of a material by applying a magnetic field in the same direction as that of the photon. By applying a magnetic field to tissue, the average plane of polarization of reflected photons can be altered; the degree of alteration depends not only on the static magnetic flux density, but also on the distance that the photon propagated in the medium. Thus,

the magnitude of rotation is a function of the average depth to which photons penetrated. There are other useful magneto-optical effects, such as the Cotton-Mouton effect, which involves a magnetic field applied to a medium perpendicular to the direction of propagation of the photons. While the photons scattered in tissue travel random directions, those that escape and that are reflected from the tissue travel, on the average, in a direction parallel to an axis perpendicular to the surface of the tissue thereby making it possible to orient a magnetic field with the average direction of photon propagation.

EXAMPLE 5

Devices

Endoscopes for cutting or puncture

It is an object of this invention to image structures, such as arteries and veins, or muscle tissue, lying below or in proximity to a region to be treated in order to prevent accidental damage to these structures. It is also an object of this invention to define an endoscopic device that can be used to image the tissues as well as puncture the membrane. In one embodiment, a retractable sheath covering a piercing device, such as a blade, is used to perform the ventriculostomy. In another embodiment, the radiant energy from an Er:YAG laser is used. The significance of Er:YAG is that the wavelength at which this operates does not penetrate the tissue to a great extent. Er:YAG is used with and without piecing elements. In yet

another embodiment, the radiant energy from an Nd:YAG picosecond microlaser is used whereupon a laser-induced plasma forms near the tissue resulting in a disruptive plasma thereby leading to perforation production.

Tissue Illuminators and Image Detectors

Illuminators are described which provide radiant energy that penetrates the skin or internal tissues. The radiant energy is either reflected or absorbed by anatomical structures, such as blood vessels, and a detector is used to analyze returning photons and the amount that is absorbed.

Because the light is absorbed and scattered to a greater extent as it travels to deep tissues, and the reflected photons reaching a particular point (x,y,z) in space are a summation of photons multiply scattered from various parts of the tissue, a means to improve contrast by discriminating against photons that do not carry significant image or spectral information is needed. Due to the small mean-free-path of more radiant energy in tissue, transilluminated images made up of unscattered photons are usually impractical, while singularly scattered light will provide little or no information since it makes up only a small part of the transmitted and reflected radiant energy and carries little imaging information.

When light from a single source is used to illuminate an object, little detail about its geometry in three dimensions is obtained without complex evaluation of the scattered and reflected radiation returning from the object. Even with complex analysis, depth perception is limited by how finely the

degrees of scatter can be calculated. To overcome this limitation, one embodiment of the present invention uses a multiple beam illuminator that emits infrared radiation that penetrates the skin from different angles. The reflected radiant energy thereby provides three-dimensional contrast as the photons move in different planes. Another device uses a transillumination scheme to achieve this same end, however in this case photons arrive at the region of interest after traveling through tissue in proximity to the object and illuminator. A combination device using both transillumination and epi-illumination; i.e., illumination from above the object, maximizes the advantages of this angular illumination.

The use of multiple illuminators, whether trans-, epi-, or in combination, also provides comparative reference points from which to subtract excess scatter which is determined by comparing rates at which photons return to a detector housed in the illuminator. Multiply scattered photons are delayed to a greater extent than those experiencing minimal scatter. These multiply scattered photons will, after being reflected to the detector, result in an image of greater distortion. Therefore, as photons become more scattered, the image becomes "fuzzy." Some of the fuzziness may be subtracted by aiming two or more illuminators at the same target and differentially measuring the length of time photons from either source return to the detector. Differentiation occurs through the use of more than one wavelength or frequency. As the distance to a known reference point is constant, there should be an optimal amount of scatter that provides a true image.

Endoscopic Visualization

In another embodiment of the instant invention, a single or multiple source illuminator is encased in a catheter or endoscopic device. The illuminator may be composed of any infrared radiant energy source but preferably is a cw or pulsed laser with fiber optic leads that results in a lens at the distal end. The laser preferably produces infrared radiant energy that penetrates through surface tissues. When combined with infrared sensing devices, the apparatus allows the endoscopic or catheter operator to visualize subanatomical structures in great detail. For example, the device allows endoscopic surgeons to visualize blood vessels that are in danger of being accidentally incised during a procedure.

Disposable endoscope and/or add-on

Because of the time, cost and sometimes ineffectiveness associated with sterilization of reusable surgical devices, it may be advantageous to produce a disposable endoscope, or part of the endoscope. The disposable endoscope, as it is not subjected to significant compressional, shear or torsional forces, is made out of thin gauge metal or even plastic. Alternatively, the majority of the length of the lumen in the endoscope is constructed of strong surgical steel, but the tip that is used to penetrate the floor of the third ventricle is made of plastic or metal and could screw into the main endoscopic body. The tip end incorporates a lens for imaging. Once used, the end is removed and disposed of, thereby improving safety and

enhancing efficiency of the fenestration process. Alternatively, a metal, plastic or rubber sheath is fixed over the entire length of the lumen of the endoscope. After use, the sheath is disposed of.

These devices incorporate a tip that is used for a surgical procedure. For example, the tip of the endoscope, whether durable or disposable, is used for puncturing or cutting the third ventricle membrane during the ventriculostomy procedure for hydrocepahalus, as described above. This tip isdesigned so as to incorporate a sharp end for cutting or puncturing. Optionally, this cutting or puncturing tip is placed on a disposable tip or sheath as well, and the disposable element discarded after the procedure so as to reduce the risk of contaminating different patients with tissue.

Microsurgical devices

Microsurgical procedures are often done through an endoscope. It is also an object of the present invention to apply these techniques and methods to microsurgical devices to provide a means for surgeons to visualize structures that may be differentiated by virtue of their infrared absorptive properties, such as blood vessels or muscle.

EXAMPLE 6

Representative configurations using endoscopic devices

Infrared sensitive CCD video with image processing and analysis system

An infrared sensitive CCD video camera (e.g., Hamamatsu C2400-79, Hamamatsu Photonic Systems, Bridgewater, NJ) and image processing and analysis system (Hamamatsu Argus-20) are coupled together. A 250 W quartz incandescent light source is optically coupled into a light transmitting fiber bundle which is coupled to an endoscope (e.g., Smith & Nephew Dyonics No. 3626S focusing video arthroscope). The proximal end of the endoscope is connected to a custom-made coherent fiber optic bundle (Schott Fiber Optics, MA) which carries the imaging information to the IR camera. Between the fiber bundle and IR camera is located an infrared long-pass filter (e.g., Edmund Scientific RG715 glass filter, Edmund Scientific, Barrington, NJ). A macrolens on a c-mount, attached to the CCD camera, is used to image the end of the fiber bundle. The image processing unit is set to enhance edges and to frame average in order to minimize the appearance of noise in the image.

Radiant energy source and imaging detector

A version of the invention consists of a source of radiant energy and imaging detector, which is configured to image subcutaneous anatomic structures. The source consists of a portable, 0.5 W continuous-wave 800 nm diode laser and power supply, while a second source consists of a 0.5 W

continuous-wave 670 nm diode laser and power supply. The power supplies are modulated on and off by a digital-delay pulse generator. The output of the lasers is collimated with lenses and configured to impinge normally on skin with a spot size of about 1 mm. The laser beams are contained within a hollow tube, which is pressed up against the skin, and which serves to contain the beams for safety reasons to shield the imaging system from the bright spots where the lasers impinge on the skin. Two CCD video cameras with imaging optics are positioned in a way such that it can image the tissue around where the lasers impinge on the skin. Each camera has a 10 nm bandpass optical filter with center wavelengths at 800 and 670 nm in front of the objective optic. The output of the laser is sent to a frame grabber and video monitor. The frame grabber is triggered by the output of the digital-delay pulse generator which is synchronized to the laser sources. The lasers are sequentially modulated on and off, and the frame grabber is triggered such that images illuminated by each laser are digitized. The frame grabber/microcomputer takes the arithmetic difference of the images in real-time such that any difference between the images is emphasized. Further real-time image processing is used to enhance contrast. A prior intravenous injection of indocyanine green would optionally enhance the contrast in blood vessels, if that is the structure to be imaged. By linearly polarizing the laser light and placing a linear polarizer in front of the camera, further contrast can be added by rotating the camera polarizer until the image improves. Optionally incorporating this imaging methodology

into an endoscope would not involve any further technology, only a decrease in size of some of the components.

Q-switched ruby laser and CCD array

Saturation of absorbing and scattering centers in tissue can be accomplished with an intense pulse of radiant energy, such as the energy emitted by a Q-switched ruby laser. The red output of this laser should be split using a beam splitter, and one of the split beams can be propagated through an optical delay line so that it recombines with the other beam, but a variable time later, for example, a few nanoseconds or so. Thus, the first pulse of energy, with a non-therapeutic energy fluence of less than 1 J/cm^2, saturates, while the second pulse a few nanoseconds later, penetrates deeper than the first pulse. The imaging system in this case consists of an intensified CCD array, electronically gated and synchronized to collect the light reflected from the first pulse and subsequently the second pulse. Various mathematical manipulations to the frame-grabbed and digitized images can be done (e.g. arithmetical subtraction) to improve image contrast.

Raman spectrograph and Nd:YAG source

A Raman spectroscopy imaging setup consists of a Raman spectrograph and Nd:YAG source, as available from Ocean Optics Inc. The output of the laser is coupled into a beam scanner, which scans the laser beam over the surface of the tissue in a raster-scanning pattern. The reflected radiant energy is collected by an optic and directed into the spectrograph. The

output of the spectrograph is coupled into an analog-to-digital converter board where the spectra are collected. The magnitude of the lines appropriate for the species of interest (hemoglobin, for example 1236 cm^{-1}), are measured and compared to the average magnitudes. A 2-dimensional map of the difference between the magnitude of the line as compared to the average is displayed on a monitor in order to provide information on the location of a blood vessel. This image is superimposed on a regular white-light image of the tissue in order to provide the health-care professional with familiar landmarks.

Photoelastic modulator and polarizing filter arrangement

A photoelastic modulator and polarizing filter arrangement, such as available from Hinds Instruments, Inc., provides a way to modulate radiant energy between different type and degrees of polarization. By directing the chopped radiant energy produced by a continuous-wave Nd:YAG laser, and setting up the photoelastic modulator/polarizer so that it changes the radiant energy circular polarization handedness sequentially between left and right, and synchronizing an imaging detector such as a CCD camera/frame grabber with the pulses of right and left hand circular polarized light, optically active species are detected.

Laser doppler flowmeter coupled to scanner

The source and collection optical fiber on a laser doppler flowmeter (available, for example, from I.S. Medtech, Inc.), is optically coupled to a scanner (available, for example. from Lincoln Laser Scanning Systems). The scanner is controlled to

scan a variable size and shape pattern on the surface of skin at a user selectable rate. The voltage output of the laser doppler flowmeter is coupled into an analog-to-digital converter board (available, for example, from National Instruments Inc.), which rapidly digitizes the voltage in synchrony with the scanning electronics. The resulting matrix of data, represented on a color or gray-scale pattern on a video monitor, serves to locate blood vessels below the surface of the skin.

Any patents or publications mentioned in this specification are indicative of the levels of those skilled in the art to which the invention pertains. These patents and publications are herein incorporated by reference to the same extent as if each individual publication was specifically and individually indicated to be incorporated by reference.

One skilled in the art will readily appreciate that the present invention is well adapted to carry out the objects and obtain the ends and advantages mentioned, as well as those inherent therein. The present examples along with the methods, devices, and specific configurations described herein are presently representative of preferred embodiments, are exemplary, and are not intended as limitations on the scope of the invention. Changes therein and other uses will occur to those skilled in the art which are encompassed within the spirit of the invention as defined by the scope of the claims.

WHAT IS CLAIMED IS:

1. A method of imaging deep anatomic structures normally invisible under white-light illumination, comprising the steps of:

inserting an endoscope into an anatomical region of interest;

illuminating said region of interest with a light source; and,

detecting infrared light from said region of interest with an infrared sensitive image detector.

2. The method of claim 1, wherein images from said infrared sensitive image detector are displayed on a video monitor.

3. The method of claim 1, wherein said infrared sensitive image detector detects infrared light reflected or emitted from said region of interest.

4. The method of claim 3 wherein said infrared sensitive image detector is selected from the group consisting of an infrared sensitive monochrome video camera, a color video camera which images sequentially through red, green, blue and infrared filters, an infrared sensitive monochrome video camera

which images sequentially through red, green, blue and infrared filters, and a color camera which is also sensitive to infrared radiation which can alternatively image through an open aperture or an infrared filter.

5. The method of claim 3, wherein said infrared sensitive image detector is a confocal imaging optical system.

6. The method of claim 3, wherein said light source is a laser which emits near infrared light of wavelengths similar to the dimensions or textures of objects being imaged such that reflection or said light results in detection of a speckle pattern.

7. The method of claim 6, wherein changes in said speckle pattern are used to detect red blood cell movement and blood vessels.

8. The method of claim 1, wherein said infrared sensitive image detector detects infrared light absorbed by said region of interest.

9. The method of claim 8, wherein differences in the absorption spectra of myoglobin, HbO2 and Hb proteins are applied determining the localization of each protein in an image.

10. The method of claim 9, comprising the further step of:

applying the localization of each protein in said image to the interpretation of said image in a manner selected from the group consisting of differentiating the location of Hb and HbO2 contained within vessels from Mb to produce good blood-tissue contrast, differentiating the location of Mb and Hb/HbO2 are used to distinguish arteries from veins, measuring the relative amounts of Hb, HbO2 and Mb to provide a measurement for ischemia.

11. The method of claim 8, wherein said detector functions as a pulse oximeter to detect blood oxygenation by absorbance of red and infrared light, wherein said light source emits red and near infrared radiant energy and said infrared sensitive image detector measures absorbance of said light.

12. The method of claim 1, wherein one or more chromophores are added to said region of interest.

13. The method of claim 12, wherein said chromophores enhance imaging by a method selected from the group consisting of adding contrast, fluorescing, and associating with specific tissues or organs.

14. The method of claim 12, wherein said chromophore is selected from calcium-linked dyes, iodine-linked agents, dye-tagged antibodies, and Indocyanine Green (ICG).

15. The method of claim 1, comprising the further step of:

treatment a region of interest with δ-aminolevulinic acid to enhancing the production of porphyrins in any malignant tissues, wherein said porphyrins act as chromophores during imaging.

16. The method of claim 1, further comprising the steps of:

transilluminating a region of interest by placing an infrared light source in contact with a tissue at or near said region of interest so that said light is scattered through said region of interest;

using infrared detectors to detect light reflected or absorbed during said transillumination to detect subsurface

structures which are differentiated on the basis of their infrared absorption or reflection properties

17. The method of claim 1, wherein depth discriminate information is obtained by illuminating said region of interest with different wavelength of light and detecting each wavelength independently.

18. The method of claim 1, wherein changes in the average range of tissue penetration of detected photons is selected by changes in the degree of detector collimation.

19. The method of claim 18, wherein detector collimation is determined by a physical collimator of a specific dimension and shape, a narrow-bandpass interferometric filter, light-absorbing baffles between objective element and detector, and confocal techniques.

20. The method of claim 1, comprising the further steps of:

saturating absorbing sites in said region of interest with a strong pulse of radiant energy;

applying additional pulses of radiant energy before the effects of prior pulses have diminished, wherein said method enhances image detection.

21. The method of claim 1, wherein detection of infrared light is enhanced by an method selected from the group consisting of Raman spectroscopy, multiphoton interaction, optical coherence tomography, time correlated single photon counting, optical rotatory dispersion, circular dichroism, polarization, chrono-coherent backscatter, simple inteferometry of backscattered light, and alignment of molecules in structures to be imaged by magnetic or electric fields.

22. An endoscopic device capable of the real time imaging subsurface structures normally invisible under white-light illumination, wherein said device comprises an endoscope, a light source, and an infrared sensitive image detector.

23. The device of claim 22 wherein said infrared sensitive image detector is selected from the group consisting of an infrared (IR) sensitive monochrome video camera, a color video camera which images sequentially through red, green, blue and infrared filters, an infrared (IR) sensitive monochrome video camera which images sequentially through red, green, blue and infrared filters, and a color camera which is also

sensitive to IR radiation which can alternatively image through an open aperture or an IR filter.

24. The device of claim 22 wherein said infrared sensitive image detector is placed on the proximal end of said endoscope.

25. The device of claim 22 wherein an image of a region-of-interest is transmitted to said infrared sensitive image detector through an image guide selected from the group consisting of a coherent optical fiber bundle and other optical image guides.

26. The device of claim 22, further comprising: a laser doppler device to detect blood flow and quantify flow rates.

27. The device of claim 22, further comprising: a transilluminator and one or more infrared detectors.

28. The device of claim 27, wherein said transilluminator is capable or transmitting light at different angles.

29. The device of claim 22, further comprising: a means of cutting or puncturing tissue on the distal end of said endoscope.

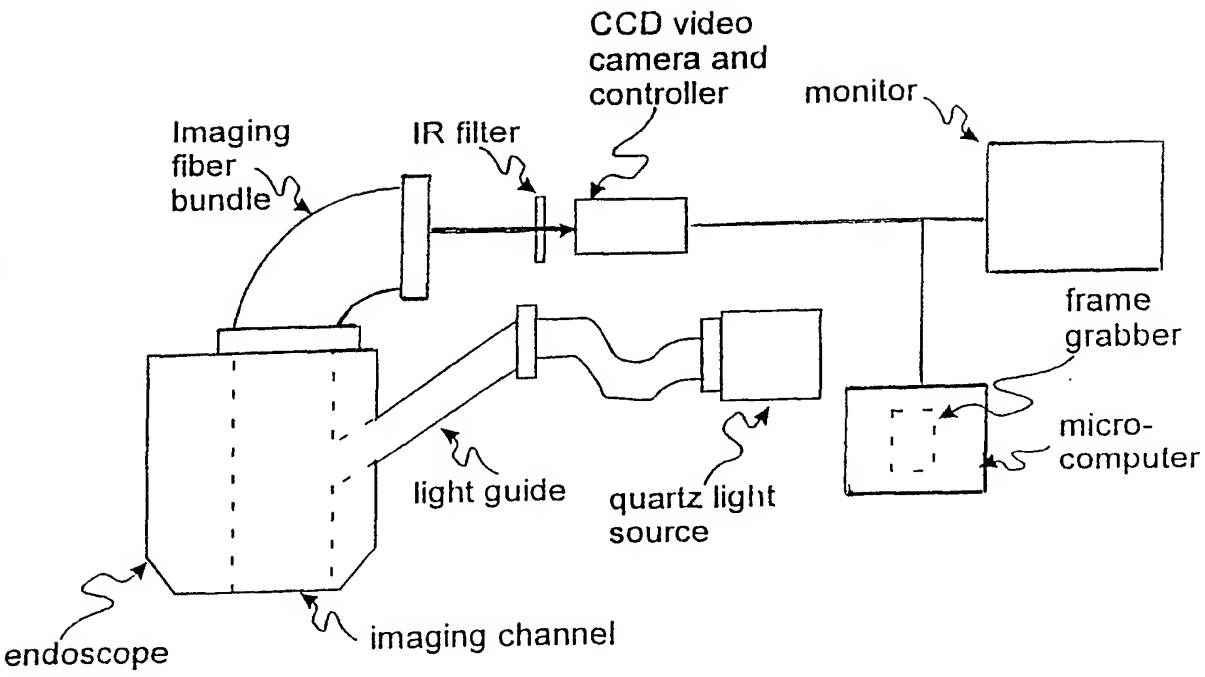

CCD video
camera and
controller
monitor
Imaging
fiber
bundle
IR filter
frame
grabber
micro-
computer
light guide
quartz light
source
endoscope
imaging channel

Fig. 1

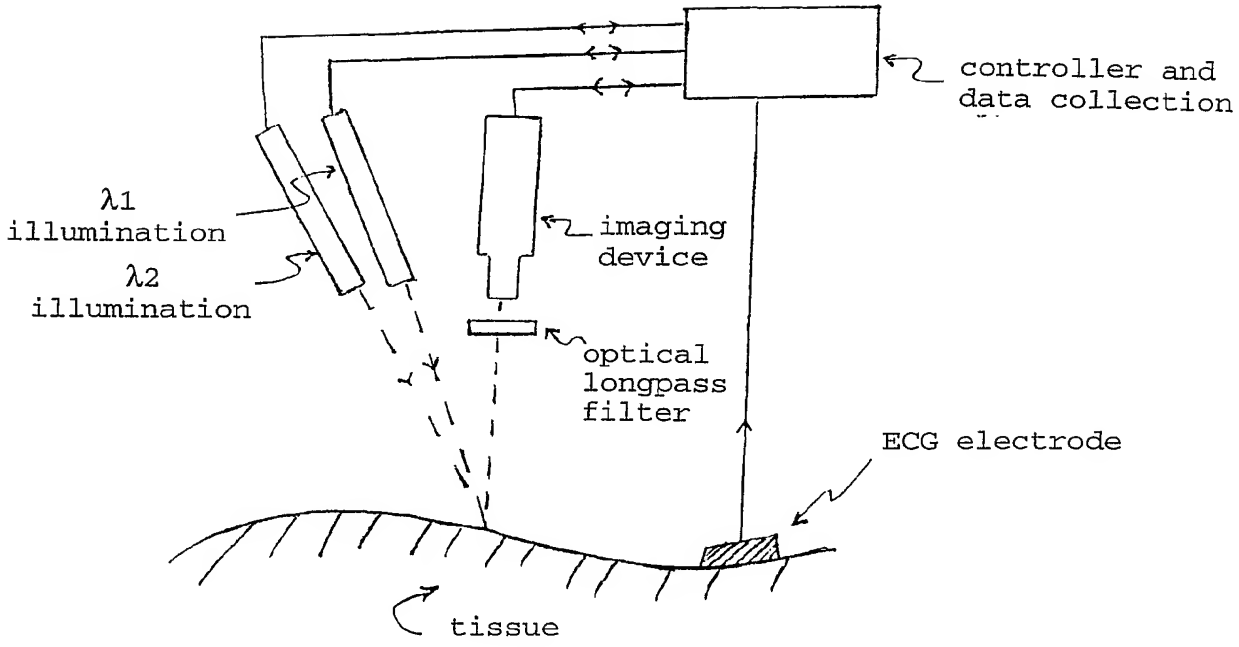

controller and
data collection
λ1
illumination
λ2
illumination
imaging
device
optical
longpass
filter
ECG electrode
tissue

Fig. 2A

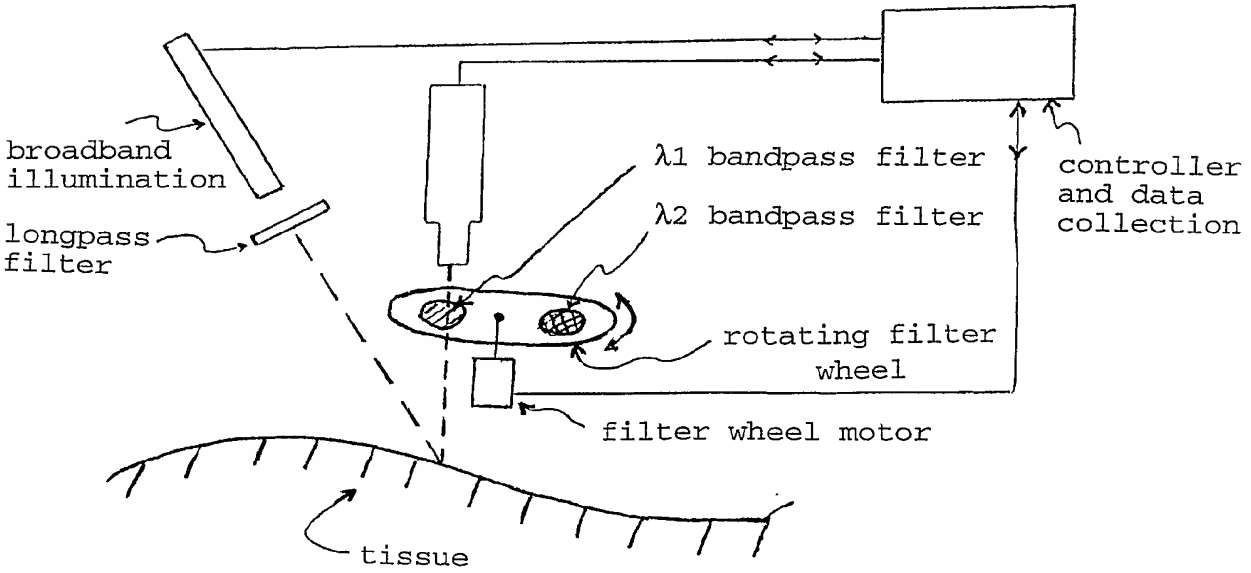

broadband
illumination
longpass
filter
λ1 bandpass filter
λ2 bandpass filter
controller
and data
collection
rotating filter
wheel
filter wheel motor
tissue

Fig. 2B

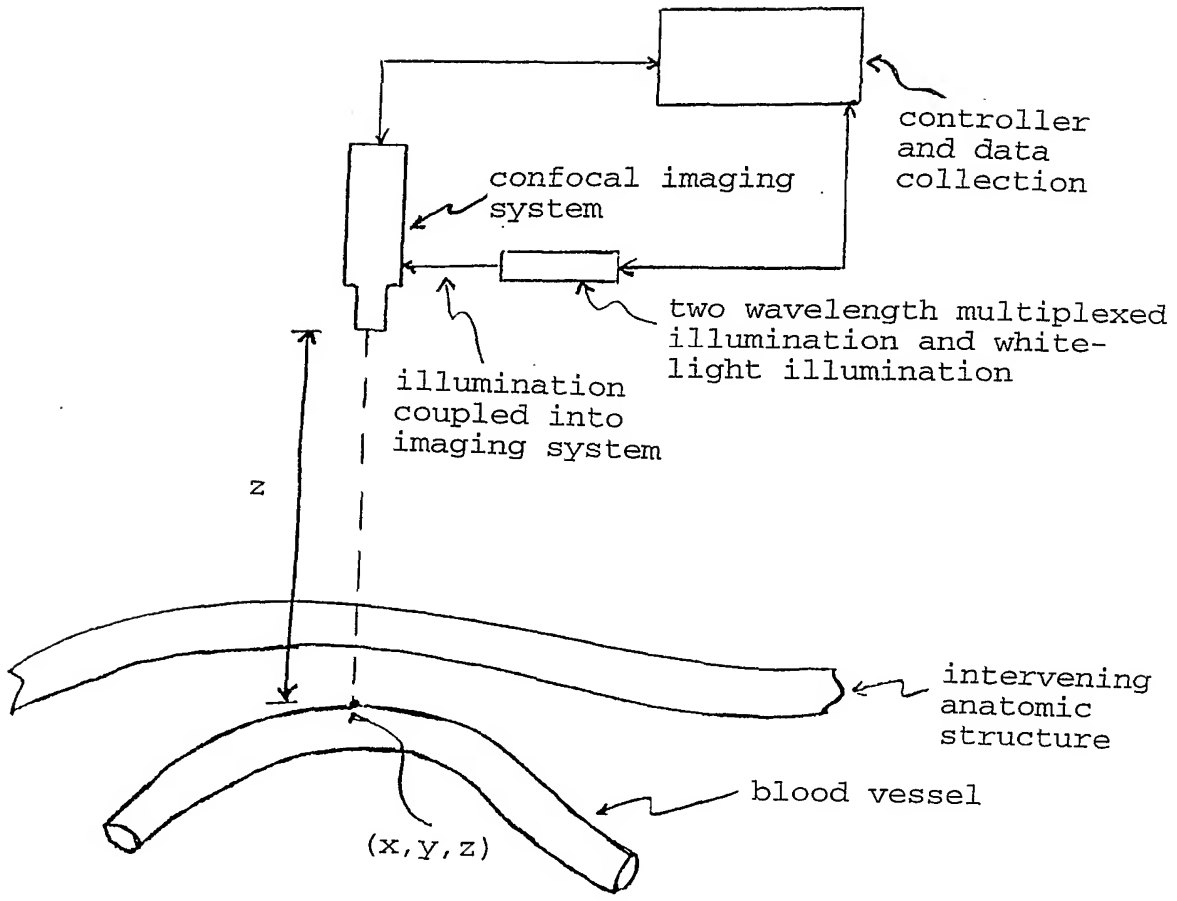

controller and data collection
confocal imaging system
two wavelength multiplexed illumination and white-light illumination
illumination coupled into imaging system
z
intervening anatomic structure
blood vessel
(x,y,z)

Fig. 3

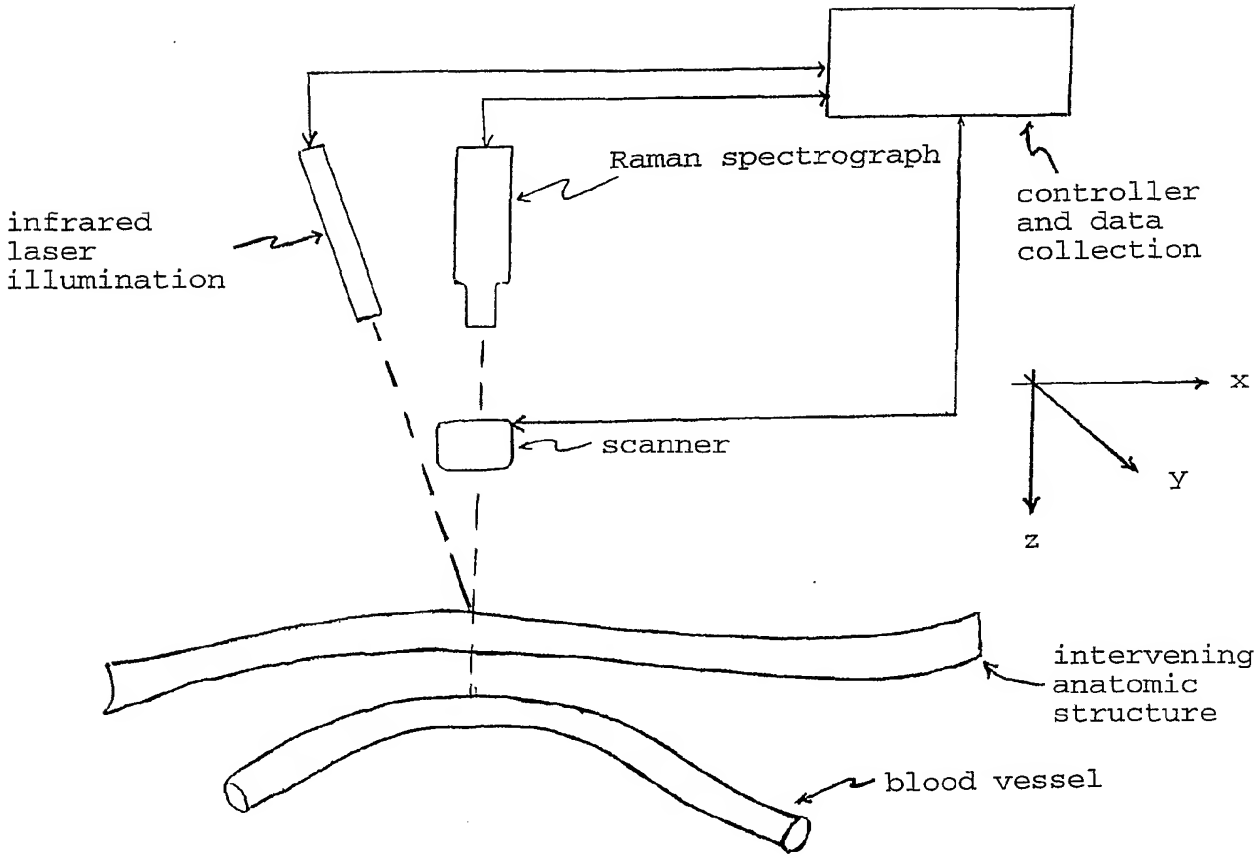

Raman spectrograph
controller
and data
collection
infrared
laser
illumination
x
y
z
scanner
intervening
anatomic
structure
blood vessel

Fig. 4

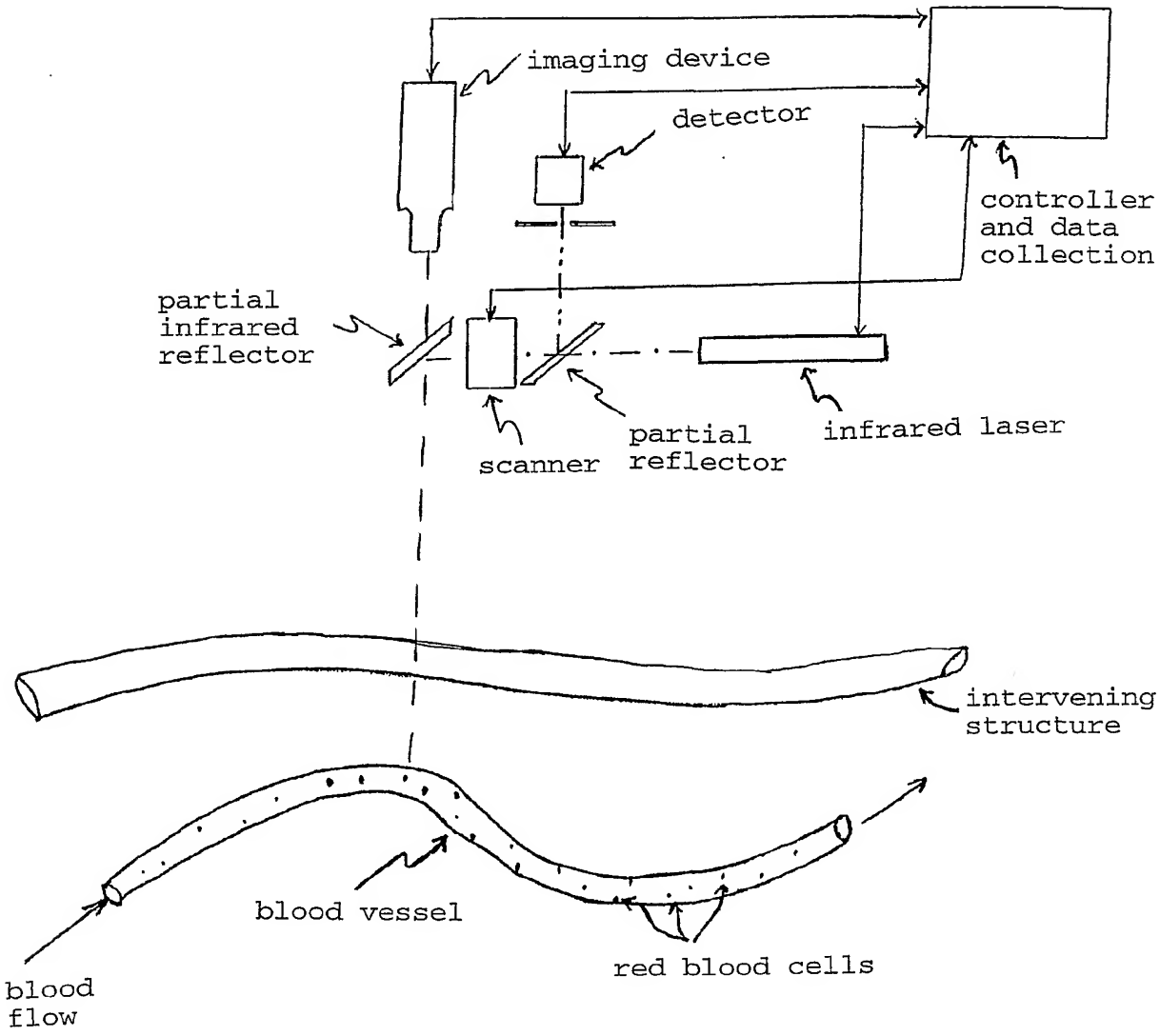

imaging device
detector
controller and data collection
partial infrared reflector
scanner
partial reflector
infrared laser
intervening structure
blood vessel
blood flow
red blood cells

Fig. 5

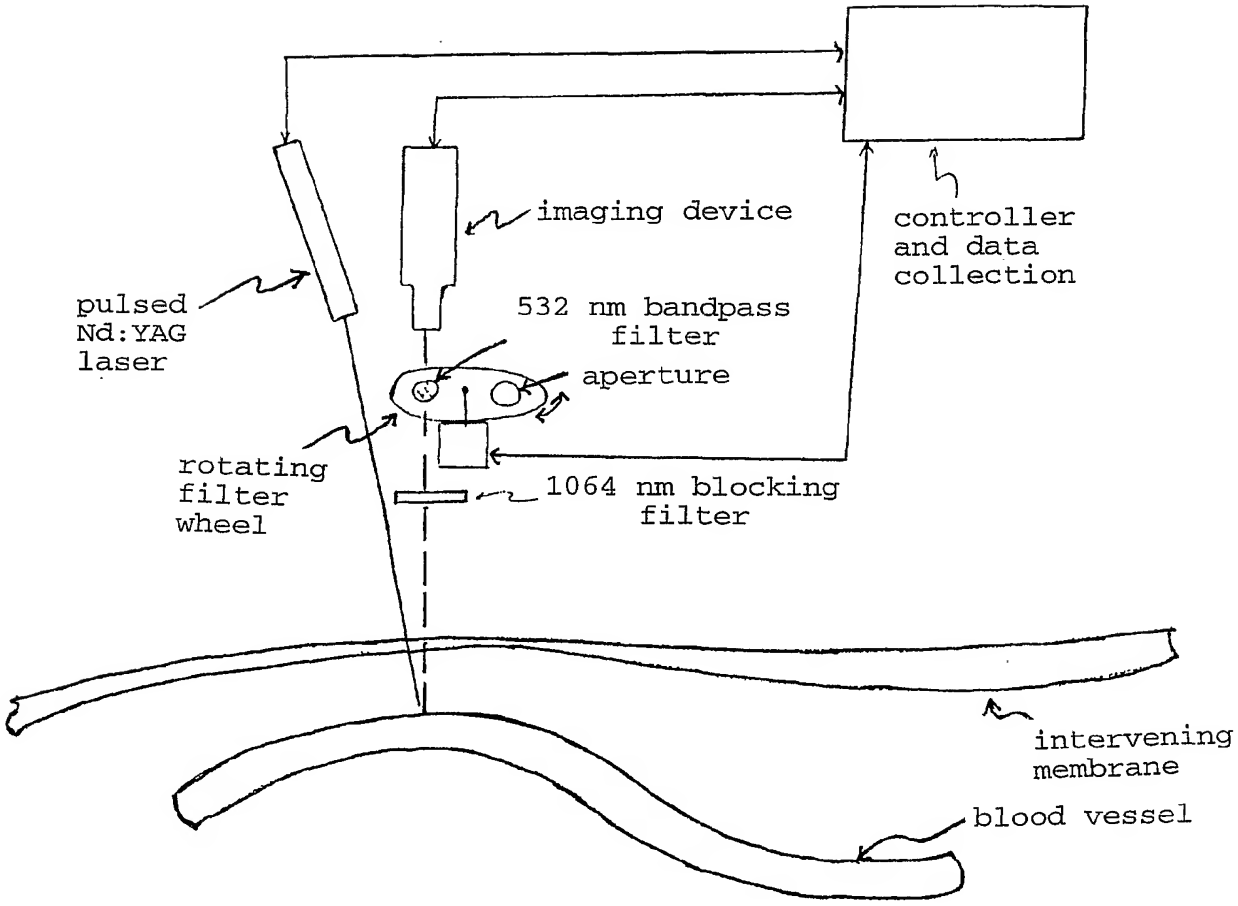

imaging device
controller and data collection
pulsed Nd:YAG laser
532 nm bandpass filter
aperture
rotating filter wheel
1064 nm blocking filter
intervening membrane
blood vessel

Fig. 6

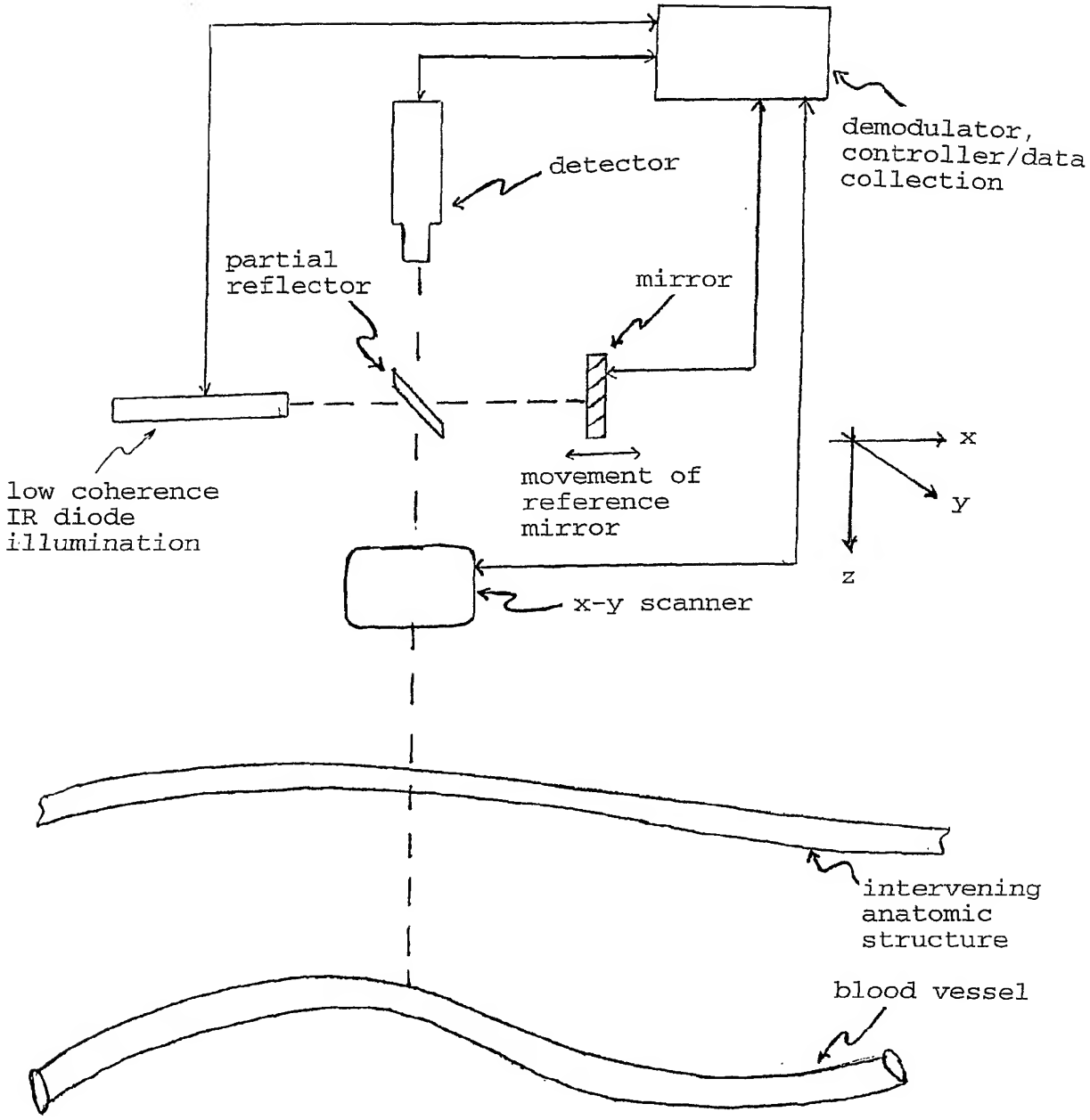

demodulator, controller/data collection
detector
partial reflector
mirror
low coherence IR diode illumination
movement of reference mirror
x
y
z
x-y scanner
intervening anatomic structure
blood vessel

Fig. 7

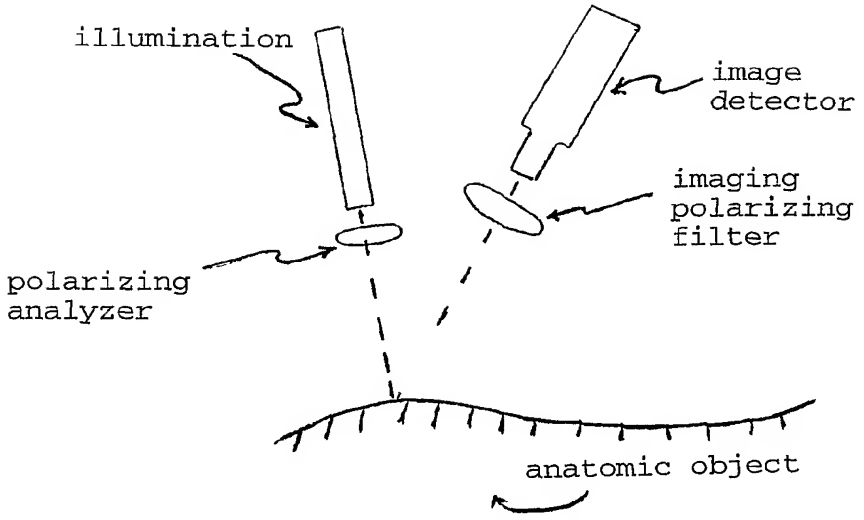

illumination
image detector
imaging polarizing filter
polarizing analyzer
anatomic object

Fig. 8A

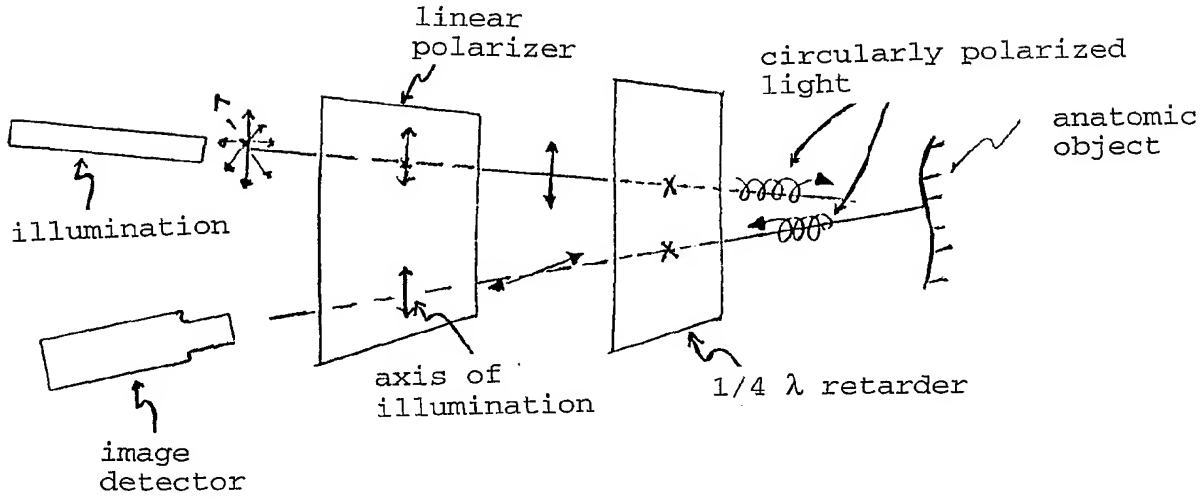

linear polarizer
circularly polarized light
anatomic object
illumination
axis of illumination
1/4 λ retarder
image detector

Fig. 8B

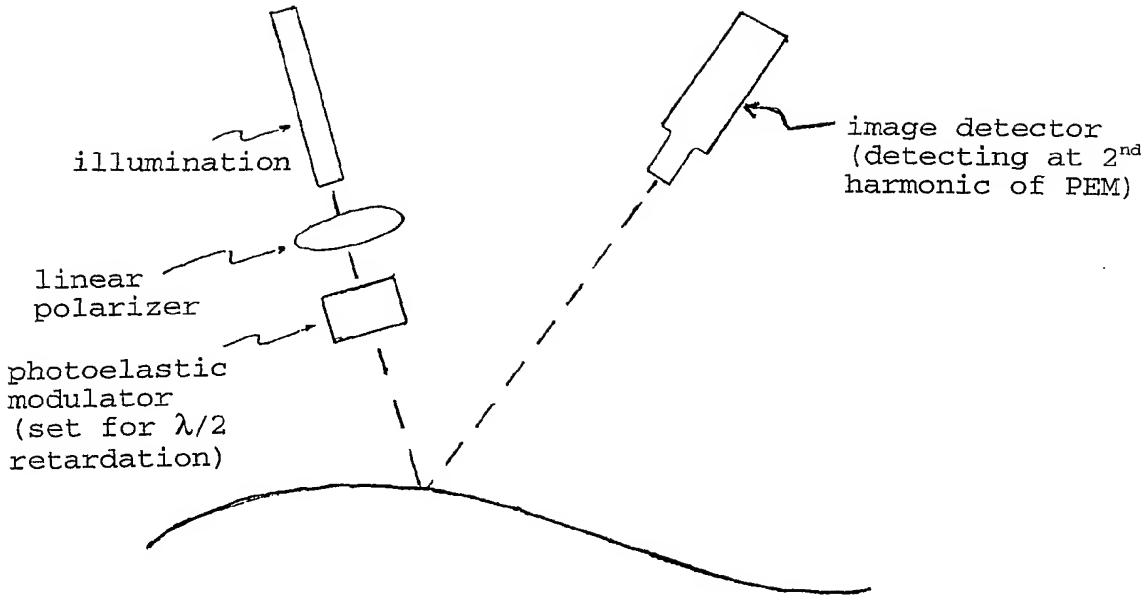

illumination
linear polarizer
photoelastic modulator (set for λ/2 retardation)
image detector (detecting at 2nd harmonic of PEM)

Fig. 8C

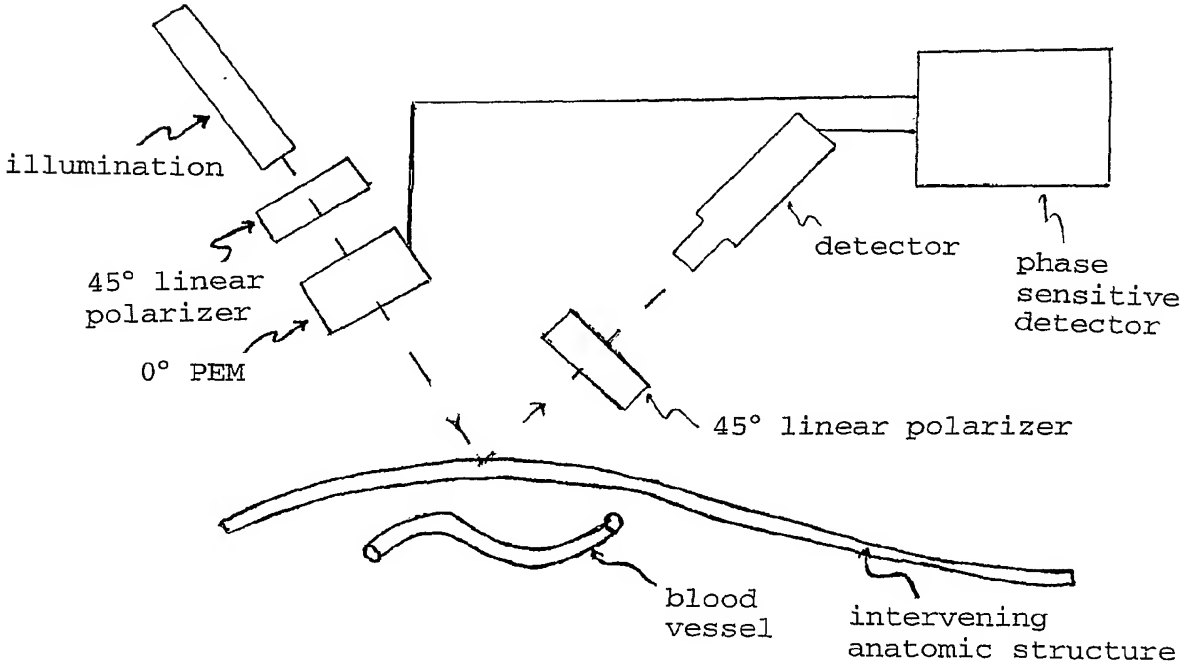

illumination
45° linear polarizer
0° PEM
detector
phase sensitive detector
45° linear polarizer
blood vessel
intervening anatomic structure

Fig. 8D

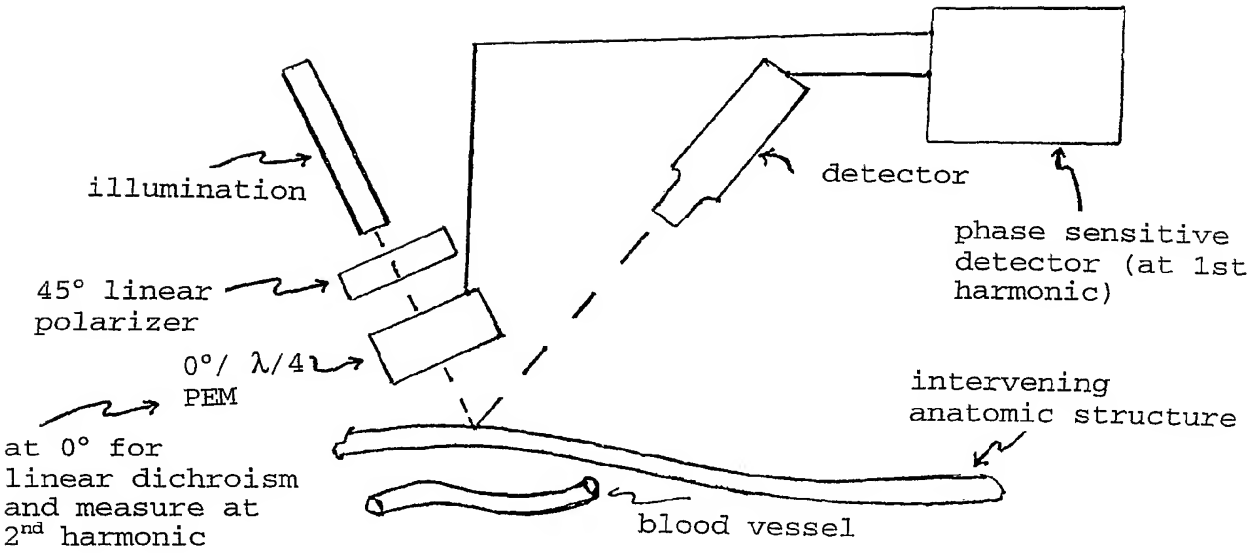

illumination
45° linear polarizer
0°/ λ/4 PEM
at 0° for linear dichroism and measure at 2nd harmonic
detector
phase sensitive detector (at 1st harmonic)
intervening anatomic structure
blood vessel

Fig. 8E

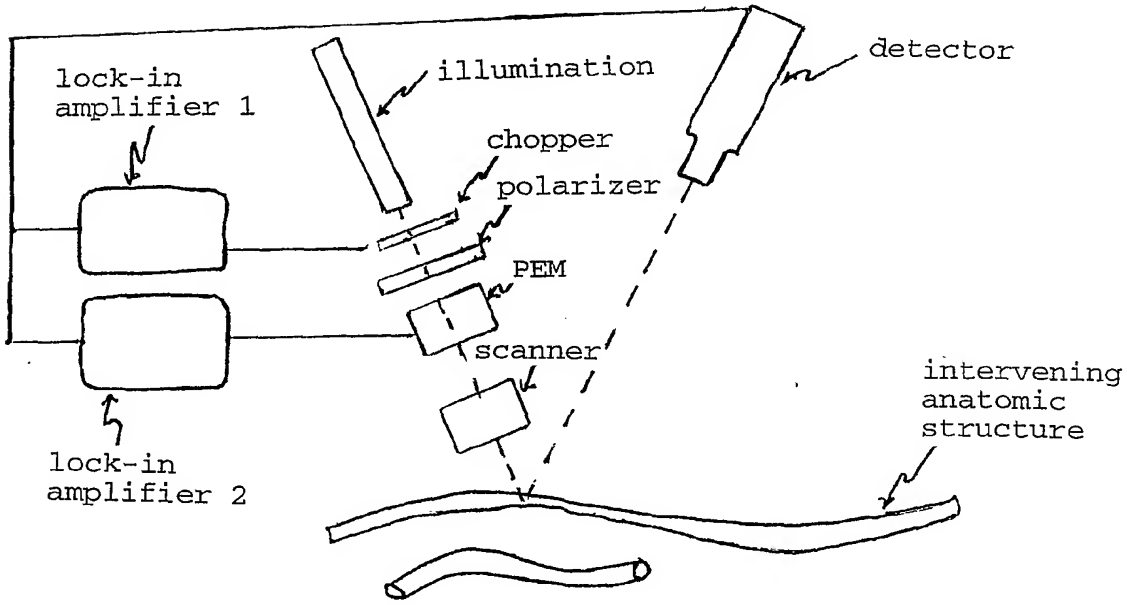

lock-in amplifier 1
illumination
detector
chopper
polarizer
PEM
scanner
intervening anatomic structure
lock-in amplifier 2

Fig. 8F

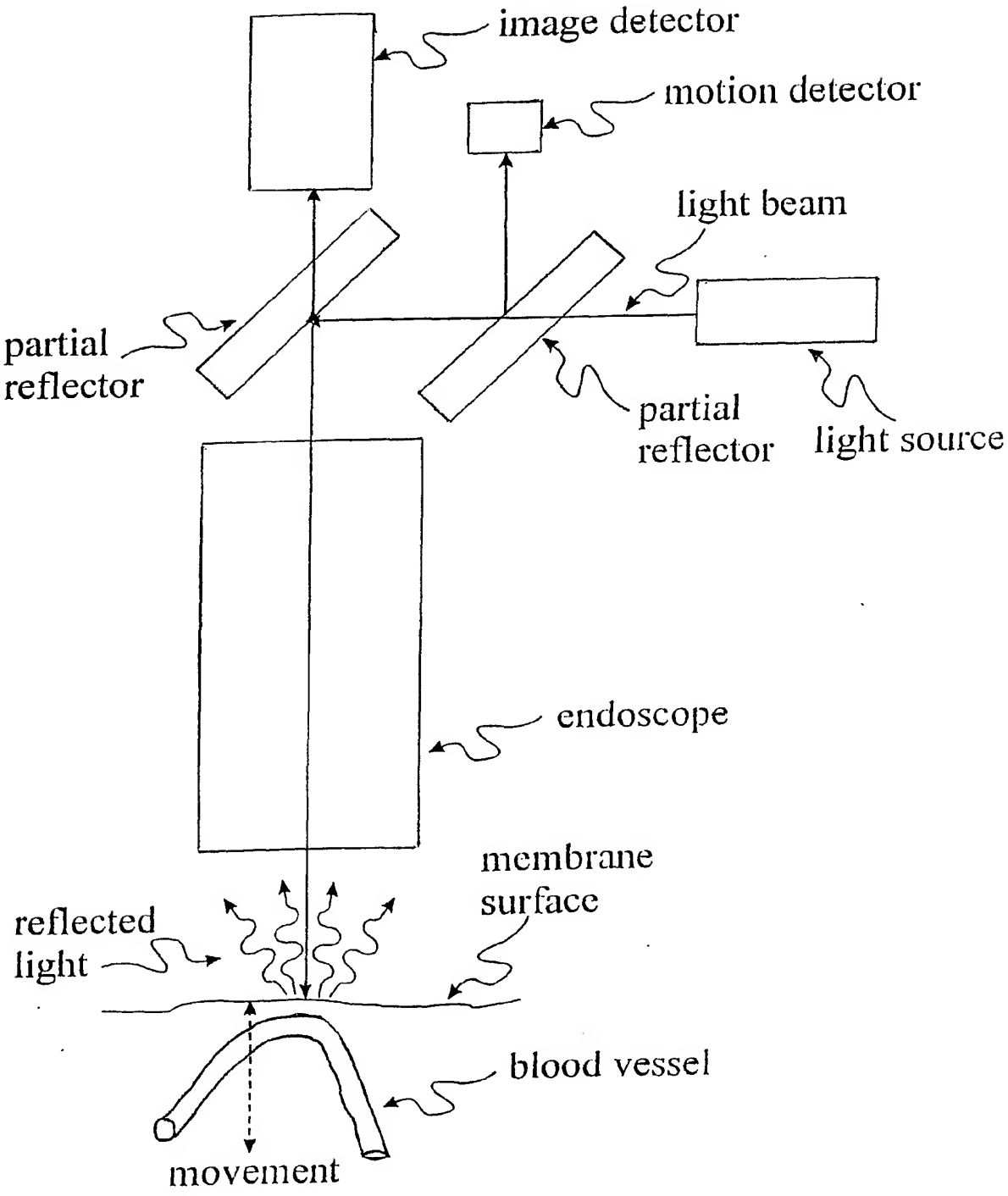


image detector
motion detector
light beam
partial reflector
partial reflector
light source
endoscope
membrane surface
reflected light
blood vessel
movement

Fig. 9

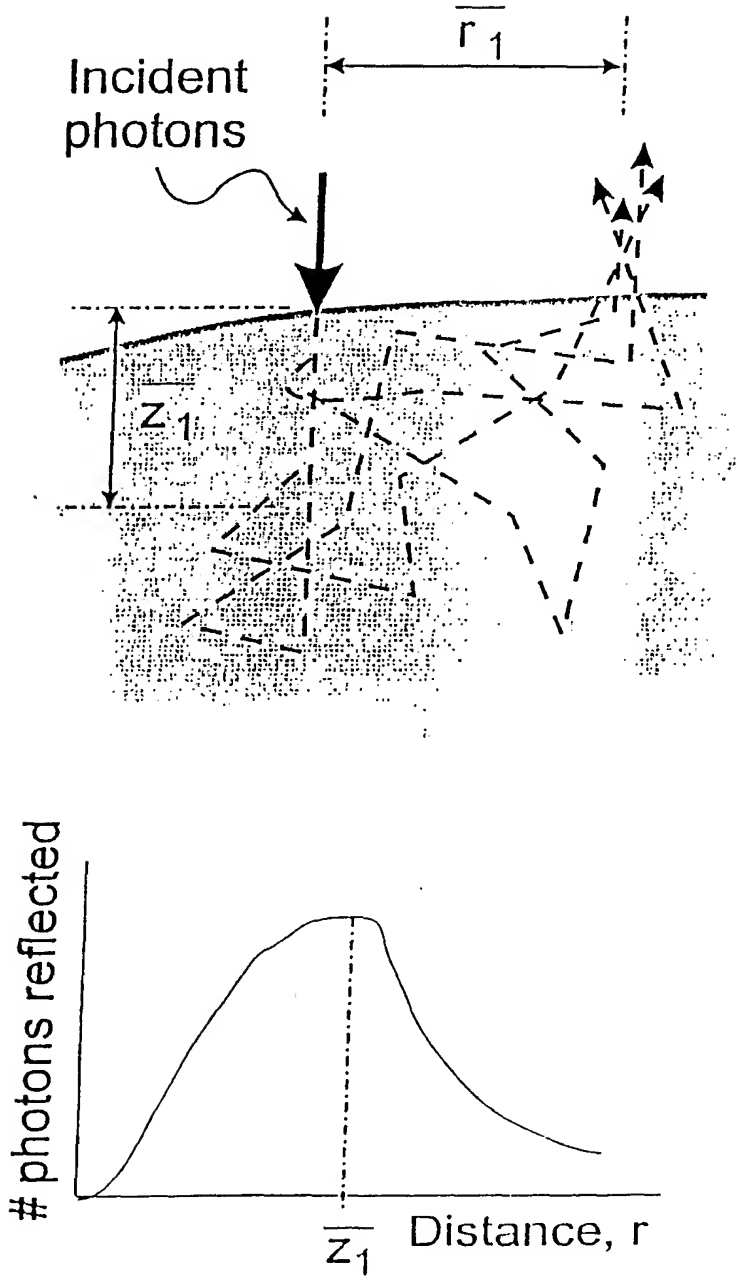
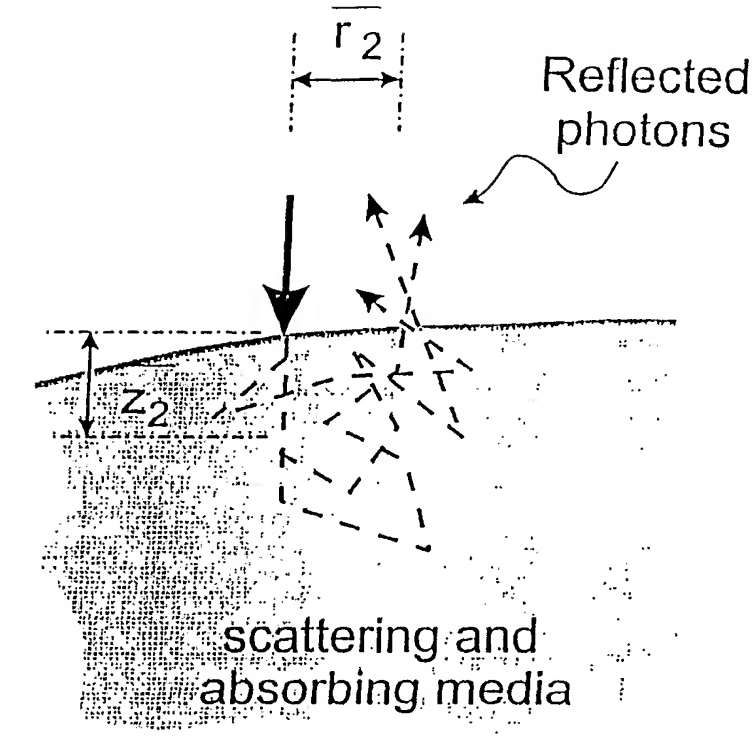

Incident photons
$\overline{r_1}$
$\overline{z_1}$
photons reflected
$\overline{z_1}$
Distance, r

Fig. 10-1


$\overline{r_2}$
Reflected photons
z_2
scattering and absorbing media

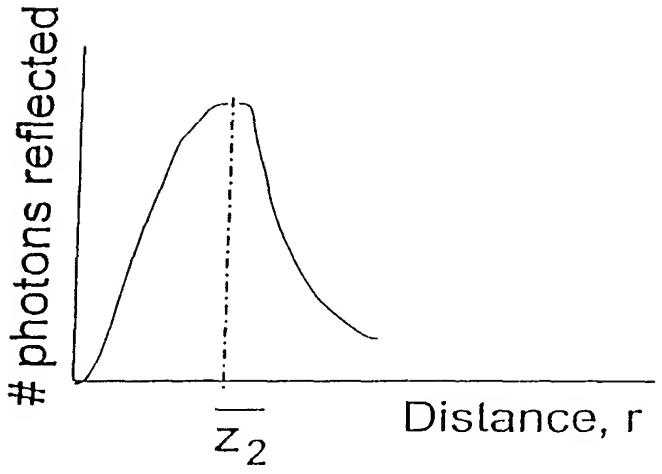

photons reflected
$\overline{z_2}$
Distance, r

Fig. 10-2

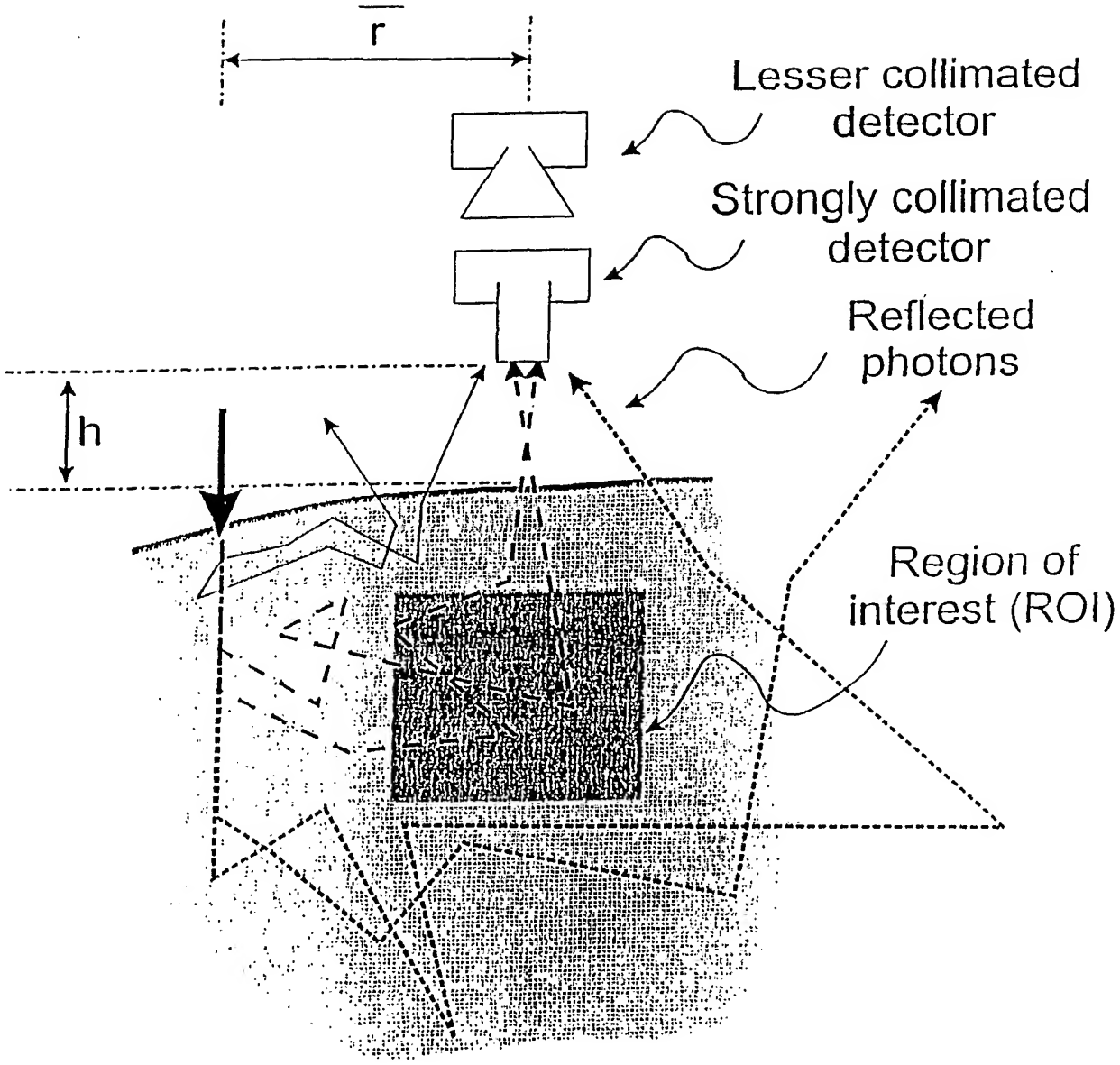

r
Lesser collimated detector
Strongly collimated detector
Reflected photons
h
Region of interest (ROI)

Fig. 11

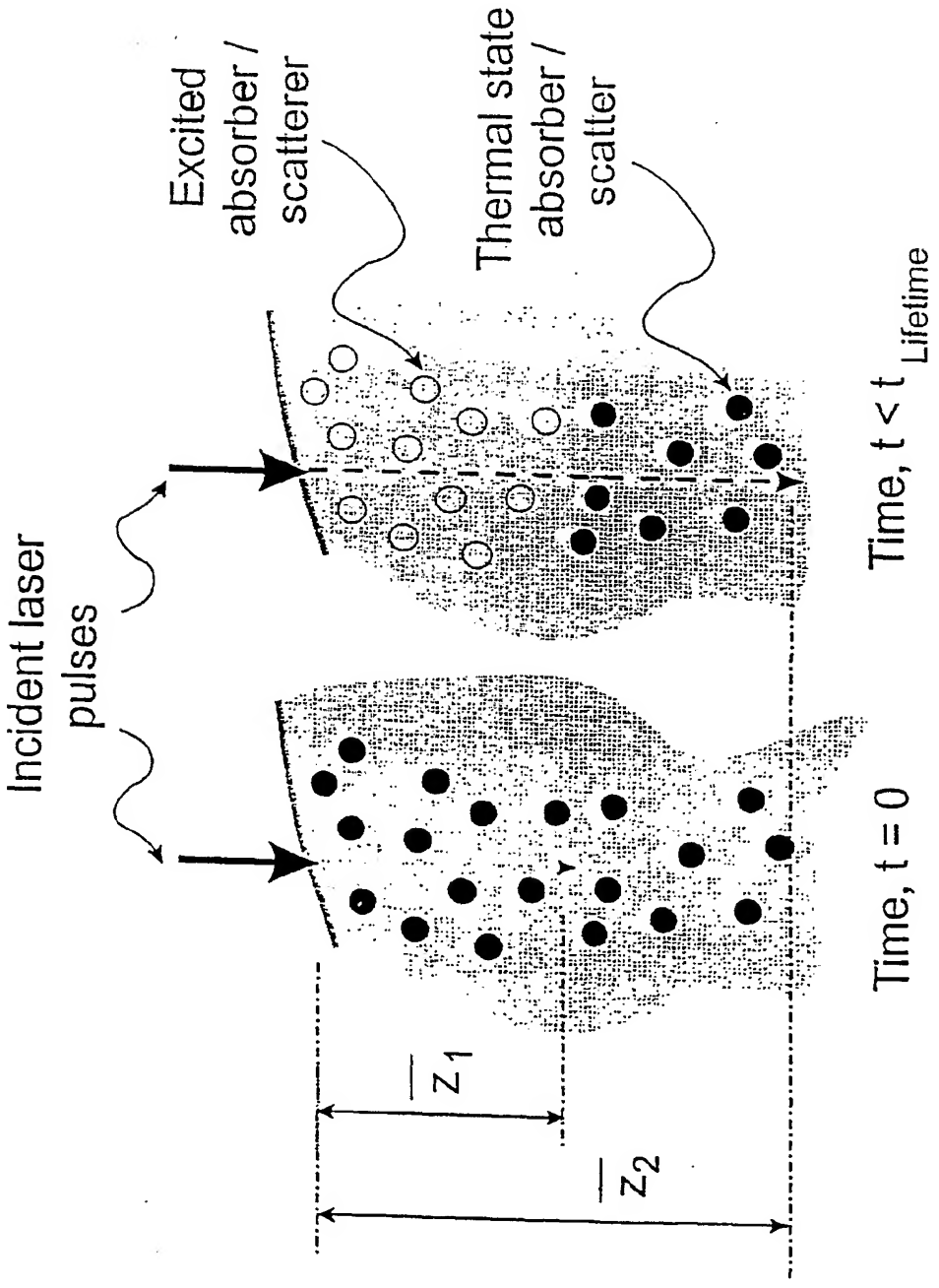

Incident laser pulses
Excited absorber / scatterer
Thermal state absorber / scatter
Time, t = 0
Time, t < t Lifetime
z1
z2

Fig. 12

INTERNATIONAL SEARCH REPORT

International application No.
PCT/US01/01159

A. CLASSIFICATION OF SUBJECT MATTER

IPC(7) : A61B 5/05
US CL : 600/473

According to International Patent Classification (IPC) or to both national classification and IPC

B. FIELDS SEARCHED

Minimum documentation searched (classification system followed by classification symbols)
U.S. : 600/473, 476, 478; 356/39, 40, 41, 42

Documentation searched other than minimum documentation to the extent that such documents are included in the fields searched

Electronic data base consulted during the international search (name of data base and, where practicable, search terms used)

C. DOCUMENTS CONSIDERED TO BE RELEVANT

Category *	Citation of document, with indication, where appropriate, of the relevant passages	Relevant to claim No.
X	US 5,845,639 A (HOCHMAN et al) 8 December 1998, see entire document.	1-29

☐ Further documents are listed in the continuation of Box C. ☐ See patent family annex.

* Special categories of cited documents:

"A" document defining the general state of the art which is not considered to be of particular relevance

"E" earlier application or patent published on or after the international filing date

"L" document which may throw doubts on priority claim(s) or which is cited to establish the publication date of another citation or other special reason (as specified)

"O" document referring to an oral disclosure, use, exhibition or other means

"P" document published prior to the international filing date but later than the priority date claimed

"T" later document published after the international filing date or priority date and not in conflict with the application but cited to understand the principle or theory underlying the invention

"X" document of particular relevance; the claimed invention cannot be considered novel or cannot be considered to involve an inventive step when the document is taken alone

"Y" document of particular relevance; the claimed invention cannot be considered to involve an inventive step when the document is combined with one or more other such documents, such combination being obvious to a person skilled in the art

"&" document member of the same patent family

Date of the actual completion of the international search	Date of mailing of the international search report
25 March 2001 (25.03.2001)	18 APR 2001
Name and mailing address of the ISA/US Commissioner of Patents and Trademarks Box PCT Washington, D.C. 20231	Authorized officer ELENI MANTIS-MERCADER
Facsimile No. (703)305-3230	Telephone No. 703 308-0858

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